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FALLOUT RADIATION EFFECTS ON LIVESTOCK (Part A) AND FOOD CROPS (Part B)

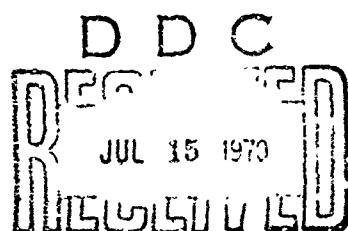
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November 1, 1968 to October 31, 1969

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UNIVERSITY OF TENNESSEE—ATOMIC ENERGY COMMISSION
AGRICULTURAL RESEARCH LABORATORY
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FALLOUT RADIATION EFFECTS ON LIVESTOCK (Part A)

by M. C. Bell, L.B. Sasser, J.L. West, and L. Wade, Jr.

AND

FOOD CROPS (Part B)

by D.D. Killion and M.J. Constantin

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Oak Ridge, Tennessee

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ABSTRACT

Sheep fed ^{90}Y -labeled fallout simulant at the rate of 2.5 mCi/kg to simulate 7% forage retention developed anorexia, diarrhea, and weight loss. Feed intake of the survivors usually returned to normal within 60 days. Beta irradiation of 57,000 rads to the dorsum of sheep, equivalent to 12% of the body surface, severely affected the skin and reduced body weight by 15% with no reduction in feed intake. Whole-body gamma irradiation of 240 R at 1 R/min affected only the platelets and white blood cells with no mortalities. These three types of irradiation given singly had little effect on mortality but when the three were combined, 4 of 8 sheep died within 60 days. In addition to mortality losses the loss of weight would severely affect the productivity of animals exposed to these levels of fallout. Preliminary data on cattle indicate that the combined radiation insults would be more detrimental to cattle than to sheep.

Gamma ray sensitivity for major crops was as follows: winter barley = winter wheat > corn > soybean > rice. Seedlings of barley and wheat will tolerate \approx 1 kR, corn \approx 2 kR, soybean \approx 4 kR, and rice \approx 25 kR. Maximum yield reduction occurred in corn irradiated from 14 to 48 DAE (days after emergence), in winter barley from 175 to 200 DAE, and in winter wheat from 150 to 200 DAE. Yield reduction in soybean was maximal at early seedling stages and during early bloom. In the monocotyledonous plants sensitivity to gamma rays is greatest during the period of apical transition and extends through the period of reproductive primordial development. In the one dicotyledonous species studied, radiosensitivity was related to the early postemergence stage and during the period of early bloom.

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F A R T A.

SIMULATED FALLOUT RADIATION EFFECTS ON LIVESTOCK

by

M. C. Bell, L. B. Sasser, J. L. West, and L. Wade, Jr.

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INTRODUCTION

There are insufficient data available to make valid predictions on the vulnerability of livestock and livestock productivity to fallout radiation in the event of nuclear war. Most of the conclusions in the literature are from data based on gamma irradiation alone (OCD-OEP-NAS Postattack Recovery, 1968). Information on ingested radioactivity is either data obtained by feeding particles to monogastric animals or from feeding soluble radioactivity to goats, dogs, and sheep. Nold, et al. (1960), fed soluble ⁹⁰Y and by calculating organ damage from dosimeter readings, indicated that the large intestine would be the critical organ. However, soluble ¹⁴⁴Ce-¹⁴⁴Pr, fed at levels lethal to 25% of the sheep, caused major damage to the omasum and rumen (Bell, 1966).

Early fallout from surface bursts is principally insoluble particulate material with a density of almost 3 but no published data are available on feeding ruminants insoluble fallout simulants at levels to cause digestive disturbances. Predictions on ingested fallout injury to livestock have been based on a low retention of fallout on forage which is not consistent with more recent data by Miller (1966).

The background information on the present research reported herein has been discussed (Bell and Cole, 1967) and a brief report of progress has been made (Bell et al., 1968). These reports demonstrate the importance of livestock as a food reserve, cattle alone being valued at over 16 billion dollars, and that the effects on livestock productivity from ingesting fallout material is potentially an important problem.

The primary objectives of this investigation were: to determine

the effects of beta irradiation to the gastrointestinal tract (GI), beta irradiation to the skin and whole-body gamma irradiation (WB). These effects were measured from single and combinations of these insults. Parameters investigated included effects on survival, feed intake, weight, blood, gross pathology and histopathology.

EXPERIMENTAL PROCEDURE

A. Sheep

Animals. To gain experience before attempting the more difficult cattle research, sheep were chosen as models since they are smaller ruminants and much easier to handle. The yearling wethers of mixed breeding used for these studies averaged 31.1 ± 0.6 kg and were randomly allotted to the 8 treatments as shown in Table 1. They were treated for internal and external parasites, and checked to insure normal blood constituents before being used in the studies. Wool was clipped from each sheep approximately one month prior to use of the animal. During a 30-day conditioning period, the sheep were gradually changed from a hay and grain ration to a daily ration of 680 g of alfalfa pellets (7 x 20 mm) plus tracemineralized salt and water. This level of feeding was selected to maintain body weight and insure complete consumption of feed. Water was also added to the pellets to reduce dusting when the radioactive sand was added. At least 7 days prior to initiation of the radiation treatments, the sheep were placed into individual stalls (Briggs and Gallup, 1949) designed for separate collection of urine and feces.

Principally due to space limitations, only 1 animal was used on each of the 8 treatments at one time. At intervals of 4 to 6 weeks,

the trials were replicated to obtain a total of 8 sheep per treatment. In addition, if sufficient fallout simulant was received, two sheep were used for dosimetry determinations.

Fallout Simulant. ^{90}Y was selected because it was readily available and has an average beta energy of 0.9 MeV, and a relatively short half-life of 64 hours. The fallout simulant was prepared at Camp Park, California through the cooperation of W. B. Lane at Stanford Research Institute and personnel of the U.S. Naval Radiological Defense Laboratory. The ^{90}Y was chemically separated from a 20 Ci ^{90}Sr "cow". Commercially available pre-sized silica sand (Wedron) was sized using a rotap to obtain sand particles between 175 and 88 μ . Sufficient sand (200 to 600 g) was added to the rotating drum of a ball mill in the hot cell and the calculated volume of ^{90}Y solution was sprayed onto the tumbling sand. The radio-tagged sand was dried and 10 ml of sodium silicate was sprayed into the rotating mixer to overcoat the particles. The synthetic fallout was transferred to a crucible and placed in a muffle furnace at 1065° C for one hour, after which it was cooled and samples taken for assay.

This insoluble fallout simulant was 99.9% free of ^{90}Sr , designed to not be absorbed from the gastrointestinal tract of sheep, and had a specific activity of approximately 10 mCi/g when fed. The half-life, energy, particle size and radioactivity of the synthetic fallout were selected to simulate fallout downwind from a one or more megaton nuclear surface burst at a distance where it is estimated that most livestock would survive gamma irradiation.

Preliminary research completed during the previous 1968 reporting

period showed that levels above 1.6 mCi/kg would consistently cause digestive disturbances in sheep. In the previous work, various levels from 0.9 to 3.2 mCi/kg were used, but in this study 2.4 mCi/kg was selected as a level which was not expected to be lethal as the only treatment. On the day that feeding was initiated, a calculated amount of sand (2.4 mCi/kg body wt) was mixed with the daily allowance of moistened alfalfa pellets. The animals were fed this mixture for 3 consecutive days. Because of the ^{90}Y decay, each animal was fed 1.85 mCi/kg the second day, and 1.44 mCi/kg on the third day.

To prevent contamination of personnel and working areas with radioactive dust, individual samples of the ^{90}Y -labeled sand were prepared inside a glove box in a hood. The radioactive sand was added to the moist alfalfa pellets in feed boxes enclosed in a large plastic bag. After gentle mixing of the feed and sand, the plastic bag was left over the feed box for at least 5 min to allow for settling of radioactive dust within the bag. During all weighing, mixing, and feeding operations, air monitors were used and all personnel wore protective gear and radiation monitors.

Skin Irradiation Sources. Eight ^{90}Sr - ^{90}Y flexible sealed sources (28 x 43 cm) were prepared (Bell, 1970). Six of these averaged about 1000 rads/hr at the surface and two averaged 1500 rads/hr. These were successfully used to provide \approx 57,000 rads to the surface of the wool on the dorsum of the sheep. Dosimetry procedures and calibrations for these plaques were described by Wade (1970). The exposure of 57,000 rads was selected to give the ratio to the 240 R of gamma exposure from information on the calculated ratio of beta to gamma on the Alamogordo

cows (Brown *et al.*, 1966).

Gamma Irradiation. Whole-body gamma irradiation was performed within 24 hours before the scheduled feeding of ⁹⁰Y-labeled sand and the beginning of skin irradiation. A facility with six ⁶⁰Co sources of \approx 12,000 Ci each was used to bilaterally expose 4 sheep at one time to 240 R at the rate of 1 R/min (C -ka *et al.*, 1970).

⁹⁰Y Excretion. Daily fecal excreta were mixed, weighed, subsampled, and oven-dried at 60° C. Dried samples were counted for bremsstrahlung using a well-type gamma scintillation counter set to exclude all pulses less than 2 MeV. This technique required a shorter decay period before counting than did beta counting and eliminated the detection of any ⁹⁰Sr contaminate. Standards were prepared by adding known quantities of ⁹⁰Y-labeled sand to nonradioactive fecal material.

Blood Studies. Blood samples were taken in heparinized tubes from which erythrocyte, hemoglobin, packed cell volume (PCV) and differential leukocyte determinations were made. Blood was drawn into a disposable pipette (Unopette) and diluted 1:1000 with ammonium oxalate for platelet determination by phase microscopy.

Periodic blood samples were taken to determine the effects of the treatments on erythrocytic uptake of ⁶⁵Zn, ⁷⁵Se, and ⁵⁴Mn (Wright and Bell, 1963) and on plasma levels of Zn, Mg, Ca, Cu, and Fe. Plasma Fe was determined colorimetrically using Ferri-Tex kits (Omnitech, Inc., Santa Monica, Calif.), and the other elements were determined by atomic absorption spectrophotometry. ⁵⁹Fe-labeled ferrous citrate was injected intravenously and its dilution in the plasma and rate of clearance from the plasma were measured. From these values the plasma volume and rate

of iron clearance (mg/day/kg) were calculated. Total body water and red cell mass were calculated from the dilution of ^3H -labeled water and ^{51}Cr -labeled erythrocytes, respectively.

Dosimetry. Radiophotoluminescent glass rods were used as dosimeters in 13 additional wethers of similar weight and age as the group described above. These rods were enclosed in threaded nylon capsules, tied together in groups of four, and sutured into place at several points of the GI tract 7 to 10 days prior to the initiation of feeding of ^{90}Y -labeled sand. Seven to ten days after feeding, at which time less than 1% of the activity remained in the animal, the sheep were sacrificed and the dosimeters removed. These same glass rods enclosed in threaded nylon capsules were used to calibrate the ^{89}Sr - ^{90}Y flexible sealed plaques as described by Wade (1970).

Pathology. Necropsies were performed on the treated animals at or near death as well as those sacrificed as controls or for dosimetry data. Primary emphasis was placed on those sheep fed radioactive sand since data are available on whole-body and skin irradiated animals. No guidelines were available on what to expect in ruminants fed an insoluble radioactive fallout simulant except for the few observations in the previous report in 1968. Gross observations were made throughout the gastrointestinal tract with special attention in the most severely affected areas of the rumen and abomasum. Ingesta was carefully washed from the mucosal surface then the affected areas were photographed if post mortem changes were not too severe. Specimens of gastrointestinal and skin tissue were collected and preserved in 10% formalin until it was processed for microscopic examination.

B. Cattle Studies

Hereford steer calves averaging a little less than 200 kg were randomly allotted to the same 8 treatments as shown for the sheep in Table 1. They were treated for parasites and checked to insure normal blood values before being used as experimental animals. In comparison with sheep, much more time was required for gentling and training before the calves could be used in the collection stalls (Hobbs *et al.*, 1950). This included stanchion feeding for 6 weeks, daily brushing for about 4 weeks and at least 8 days in the collection stalls before initiation of the treatments.

During the training period, the steers were gradually changed from a ration of hay and grain mixture to the pelleted alfalfa used in the sheep studies. Tracemineralized salt was also fed to the steers during the training and experimental periods. Daily intake of alfalfa pellets was restricted to 2.7 kg to insure complete consumption of feed during the time the steers were in the collection stalls. The ^{90}Y -labeled sand was added to the moistened alfalfa pellets to provide 2 mCi/kg of body weight for 3 consecutive days. Two sealed ^{90}Sr - ^{90}Y plaques were used on the dorsum of each steer to irradiate approximately 8% of the body surface. All other procedures were essentially the same as those listed for sheep except that the less important observations were omitted.

RESULTS

A. Sheep

Mortality. The feeding of ^{90}Y -labeled sand at the rate of 2.4 mCi/kg was a major contributor to the early deaths, especially in combination

with whole-body gamma irradiation (Table 1). Both GI and skin irradiation contributed to the deaths beyond 61 days. Pathological observations on the sequential development of the GI lesions will be discussed in detail below.

Gross Pathology. The response of various tissues and/or organs, particularly the gastrointestinal (GI) tract of sheep, exposed to the types of irradiation indicated, can probably best be appreciated by considering them in a sequential or developmental order. The usual individual biologic variation in responses was apparent. The day listed refers to the interval between the first day of treatment and necropsy. The number in parenthesis indicates the number of animals examined on that day. Unless otherwise stated, all sheep were fed 2.4 mCi $^{30}\text{Y}/\text{kg}$ of body weight.

a. Gastrointestinal Irradiation: Most GI lesions were found on the floor or ventral aspect of the rumen or the floor and ventromedial or ventrolateral aspects of the wall. Most reticular lesions were observed on the ventral aspect or floor of the organ. Lesions in the omasum occupied the ventral or free aspect of the lamina, usually the anterior portion. Abomasal lesions usually occurred in the caudal fundus and cephalic pylorus on the greater curvature. There were no lesions which could be attributed to treatment in sheep examined on day 2 (1) and 3 (2).

Day 4 (2). Several small 1 cm polyp-like nodules were observed in the ruminal sac cephalic to the posterior pillar of one sheep, but no lesions were evident in the rumen of a second sheep. The abomasal mucosa of both were slightly edematous and hyperemic which was similar to

changes present in sheep examined day 2 and 3 and also were observed in 2 of 7 presumably normal control sheep.

Day 5 (1). A 3 x 4 cm tan-colored polyp-like nodule in the ruminal sac and 5 soft fluctuating nodules in the posterior blind sac were present.

Day 7 (2). A 10 x 7 cm area of hemorrhage and an area of fibrino-necrosis with small polyp-like nodules at the periphery were seen in the anterior ruminal sac. Papillae surrounding the necrotic area were enlarged and some were apparently confluent or coalesced. There was a yellowish gelatinous exudate over the serous surfaces at the ruminal reticular junction. Small hematomas were observed at the ventral-anterior aspect of the free borders of a few major omasal laminae.

Day 8 (4). Clusters of tan-colored polyp-like nodules (0.5 to 5 cm) were observed in all ruminal compartments. The surface of some nodules was smooth, while others were covered with papillae. There were areas of depigmentation and depapillation. In some areas over the mucosal lesion the entire thickness of the ruminal wall was involved with an inflammatory reaction. A small flat sessile-based yellowish nodule was adherent to the reticular mucosa. There were linear streaks of necrosis on the free aspect of a few major omasal laminae. Abomasal changes varied from moderate edema and hyperemia to linear hemorrhagic necrosis involving the mucosa of the caudal fundus.

Day 9 (2). Large areas of fibrino-necrosis studded with polyp-like nodules, large smooth surfaced pedunculated tan-colored mass and a large dome-shaped mass covered with papillae had developed in the ruminal sac. The surrounding papillae were enlarged and some appeared to have

coalesced. Serosal adhesions to surrounding structures occurred over some lesions. Abomasal changes consisted of a large area of hemorrhagic necrosis involving several laminae in the caudal fundus and the cephalic pylorus.

Day 11 (4). Elevated yellowish-tan plaques of fibrino-necrosis were observed in the ruminal sacs. These plaques were studded with polyp-like nodules in many instances. The surface of some nodules was smooth while others were covered with necrosing papillae. The edges of some plaques were detaching, revealing a smooth pale base. A few yellowish smooth, sessile-based nodules were firmly adherent to the mucosa of the reticulum. Omasal changes varied from superficial erosions and hyperemia to small fibronecrotic nodules attached to the free edge of two major laminae. Abomasal changes varied from a small area of superficial necrosis at the crest of one lamina to areas of hemorrhagic necrosis (2, 2.5 and 4 cm) involving principally the caudal fundus and the cephalic pylorus to a lesser extent. Caudal fundic laminae in the larger necrotic area were sloughing.

Day 12 (2). Ruminal changes were similar to those of Day 11. Several tan-colored, sessile-based, dome-shaped nodules were adherent to the walls and floor of the mucosa of the reticulum. An area of hemorrhagic necrosis (4 x 3 cm) involved 5 laminae of the caudal fundus of the abomasum of 1 sheep. The reaction extended to and involved the serosa.

Day 13 (1). One small area of hemorrhagic necrosis and two areas (6 x 6 and 2.5 x 1 cm) of elevated yellowish fibrinonecrosis in the form of polyp-like nodules were observed in the rumen. There were

streak subserous hemorrhages and the serosa was adherent to adjacent structures. A small area of hemorrhagic necrosis involved the caudal fundus of the abomasum. The fundic mucosa was markedly edematous and hyperemic. This sheep had received 1.6 mCi of ⁹⁰Y-labeled sand/kg of body weight for 3 consecutive days.

Day 15 (1). Areas of yellowish fibrino necrosis were observed in all ruminal compartments (7×4.5 , 15.5×7 , 3.5×2.5 cm). The lesions were detaching at the edges. The inflammatory reaction involved the entire ruminal wall over the largest lesion. The abomasal mucosa was slightly hyperemic and edematous. This sheep had ingested 1.6 mCi ⁹⁰Y-labeled sand/kg body weight for 3 consecutive days.

Day 19 (1). A Y-shaped hemorrhagic scar was observed in the ruminal sac, one limb (5×1.5 cm) was covered with a tan-colored necrotic exudate. In another compartment were two tan-colored necrotic plaques (13×7 , 4.5×3.5 cm). The edges of the plaques were detaching, revealing a hemorrhagic base. An area of hemorrhagic necrosis involved the caudal 5 cm of the fundic and 3 cm of the cephalic pyloric abomasal mucosa. The mucosa was hemorrhagic and edematous. The medial aspect of the abomasum was adherent to the rumen.

Day 20 (1). Large areas of fibrino-necrosis (5.5×4 , 4.5×4.5 , and 10×8 cm) affected the ruminal compartments. The necrotic masses were friable, detaching, rolling up at the edges, and the underlying base was smooth and pale. Serosal adhesions had developed over the lesions. The caudal fundic laminae were thickened, hyperemic, and were studded with several ecchymotic hemorrhages.

Day 23 (1). A pale 12.5×2.5 cm scar and a 6.5×3.5 cm area of

yellowish-green fibrino-necrosis was observed in the anterior ruminal sac. A few small polyp-like nodules of yellowish fibrino-necrosis studded the posterior ruminal sac. A 7.5 x 2.5 cm area of yellowish-red fibrino-necrosis involved the caudal fundic region of the abomasum. The mucosa was dull red in color and moderately edematous. The abomasum was adherent to the omasum. There was a moderate degree of sanguineous ascitic fluid and an excessive quantity of clear fluid in the thoracic cavity. The cardiac chambers were dilated. The lungs were heavy and reddish-gray in color.

Day 59 (1). Dark-colored stellate scars (6.5 x 5, 10 x 7, 5 x 4 cm) were present in the ruminal sacs. Yellowish necrotic tags were adherent to the largest scar. A 10 x 7 cm area of the caudal fundic and cephalic pyloric abomasum was firmly adherent to the abdominal wall. An ovoid-shaped ulcer (4 x 2.5 cm) with sharply demarcated borders was partially filled with a yellowish caseous plug. A few caudal fundic laminae adjacent to the ulcer were necrotic. The skin overlying the adhesion was cyanotic, indurated, and thickened. An abomasal fistula or hernia conceivably would have developed if the sheep had survived for a longer period.

Day 68 (1). An epilated soft fluctuating pendulant enlargement (6.5 x 4 cm) anterior to the prepuce was observed 68 days following cessation of treatment. Ruminal scars were observed in the various ruminal compartments (10 cm, 2.5 x 2 cm and a 7 x 6 cm scar with a 2 cm ulcer covered with caseous exudate). Serosal adhesions to the abomasum had developed. Yellowish gelatinous exudate covered areas of the rumen and abomasum. A 6 x 5.5 cm ovoid diverticulum involved the caudal

fundus of the abomasum on the greater curvature. The periphery of the diverticulum was firmly adherent to a hernial ring in the abdominal wall. The surface of the diverticulum was covered with a dirty yellowish necrotic exudate. A 2.8 cm healed scar extended from the diverticulum into the cephalic pylorus.

Day 83 (1). Portions of the caudal fundus and cephalic pylorus of the abomasum prolapsed through a hernial ring in the abdominal wall. The prolapsed tissue was markedly edematous, hyperemic and oozed sanguineous fluid. About 7 cm of the caudal fundus was firmly adherent to the wall of a 6 to 7 cm hernial ring in the abdominal wall. Stellate and linear scars (10 x 3, 11 x 7, 4 x 3 cm) were observed in the ruminal compartments. The center of the largest scar was studded with several necrotic areas and serosal adhesions occurred over the lesions.

Day 134 (1). A rumen fistula about 1.2 cm anterior to the prepuce was observed to occasionally discharge greenish colored fluid from the tract. A fistulous tract 2.5 x 1.5 cm and about 3.5 cm in length had developed just anterior to the pillar and communicated with the exterior. A penetrating ulcer occupied the posterior portion of the erosion in the caudal ventral ruminal sac. An 8 x 1.5 cm deep erosion had developed over the posterior pillar. The anterior sac was marked by a healed 7 x 1.5 cm scar. A 3-cm linear fissure occupied the caudal fundus and cephalic pylorus of the abomasal mucosa. This lesion was not associated with an inflammatory reaction or exudation.

Day 171 (1). An abomasal prolapse involving about 7 cm of the caudal fundus and 9 cm of the cephalic pylorus developed. About 8 cm of the caudal fundus was firmly adherent to the wall of a 10 cm hernial

ring. The abomasal tissues were markedly edematous, hyperemic and thickened. A healed 11 cm scar was observed in the anterior ruminal sac. A pale 6 cm area studded with small papillae and an 8 x 2.5 cm scar with linear erosions which were covered with yellow-gray necrotic exudate were present in the ruminal and posterior ventral sacs, respectively.

Day 203 (1). An abomasal prolapse involving about 9 cm of the caudal fundus and the greater part of the pylorus developed. The prolapsed tissue was deep red in color, markedly edematous and the surface was studded with numerous gray foci. The caudal fundus was adherent to a small hernial ring (2.5 cm). The laminae in the region were edematous and congested. There were several superficial areas of mucosal necrosis. The proximal duodenal mucosa was congested. A few superficial necrotic foci were observed. The various ruminal compartments were marked with scars (14 x 3, 4.5 x 1.5, 1.5 x 1 cm). The centers of the smaller scars were studded with necrotic areas.

Days 309 (1), 346 (1), and 374 (1). Several scars varying from 2.5 to 14.5 cm were observed in the ruminal compartments. Foci of necrosis, with or without caseous exudate, involved the surface of some scars. The size of the scar was not related to the presence or absence of these reactive foci. Abomasal scars involving the caudal fundus and cephalic pylorus were present in two animals. The scars were from 6 cm in length by 2 cm in width and involved the entire wall.

Day 387 (2). There were no GI lesions in one sheep fed 1 mCi/kg body weight. Another animal with identical treatment had three ruminal scars (4 x 2 cm) and associated serosal adhesions. There was no evidence

of abomasal injury. A third sheep fed 2 mCi/kg of body weight for 3 days and examined 330 days later showed scars in the ruminal compartments (10×2.5 , a Y-shaped $5 \times 3 \times 3.5$, 4.5 cm). There was a 7-cm scar involving the caudal fundus and cephalic pylorus of the abomasum.

b. Combined GI and WB Irradiation:

Day 4 (1). No ruminal changes were observed. An acute hemorrhagic abomasitis involving the caudal fundus and cephalic pylorus on the greater curvature was apparent. Pericardial and peritoneal fluids were increased. The cardiac ventricles were dilated. The lungs were distended, heavy, firm on section and red-gray in color.

Day 17 (1). Yellowish, elevated, polyp-like nodules of fibrin-necrosis (6×5 , 4.5×3 , 5×2.5 cm) were observed in the ruminal compartments. Serosal adhesions to surrounding structures were present over some lesions. A large area of hemorrhagic necrosis (11×5.5 cm) involved the caudal fundus and cephalic pylorus of the abomasal mucosa. Some laminae of the caudal fundus were necrotic and sloughing. There was a fibrino-hemorrhagic serosal adhesion to the abdominal wall. A frothy, slightly purulent exudate was observed in the bronchi and larger bronchioles. The right lung was distended and grayish in color. All cardiac chambers were filled with clotted blood.

c. Skin-plaque Irradiation: In general the dorsum of the sheep irradiated with the sealed ^{90}Sr - ^{90}Y sources appeared to show the progressive changes reviewed by Conard (1956). These included cessation of wool growth, excess moisture and heat loss and eventual fibrin exudate with sloughing of the debris. In addition, the back was very sensitive within a few days with some evidence of locomotor disturbances

and/or incoordination of the posterior quarters. In some of the skin-irradiated sheep this became progressively worse until they were unable to use their hind limbs.

Day 54 (1). A moderate degree of hydropericardium was present. The lungs were reddish-gray in color, less crepitant and sectioned easily. The abomasal mucosa was hyperemic. The irradiated skin was intact, moderately pliable, somewhat thickened, and dark reddish-blue in color. There were areas of epilation.

Day 103 (1). Changes consisted of slight ascites and hydropericardium. Pulmonary air passages contained frothy fluid.

Day 123 (1). Large areas of desiccated serum coagula was loosely adherent to wool fibers in some areas. The surface of epilated areas was covered with dark brownish-black coagula. The underlying tissue was firm, inelastic, and dark colored. Some serum scabs covered pools of purulent exudate. There were areas of fibrotic proliferations in the deeper derma and subcutaneous tissue.

d. Combined Whole-body (WB) and Skin Plaque Irradiation:

Day 158 (1). This animal was unable to stand and was destroyed. The cardiac ventricles were dilated and the papillary muscles of the left ventricle were enlarged and gritty on section. The dermis and subcutis appeared to be more firmly adherent to the vertebral ligaments than normal. Areas of epilation alternated with areas covered with wool in the irradiated thoraco-lumbar area. Serum-blood coagula was adherent to wool fibers. The tissue was firm and inelastic. The dermis and subcutis were thickened. There were numerous grayish-white streaks in the longissimus dorsi muscle.

Day 242 (1). The lungs were heavy, edematous, reddish-purple in color and non-crepitant. There was a moderate degree of sanguineous pericardial fluid but the heart chambers were virtually empty. The surface of the irradiated skin area was congested, hemorrhagic and large areas were covered with an exudate presumed to be composed of serum coagula and necrotic material. A grayish-white slightly elevated band marked the junction of the irradiated and nonirradiated skin. The underlying muscle was pale, soft, moist, and marked with grayish-white striae. The subcutaneous tissue over the gluteal region contained an inflammatory exudate.

e. Combined GI, WB, and Skin Plaque Irradiation:

Day 21 (1). Large areas (8×5 , 4.5×1 , 13×8.5 , and 3×3 cm) of fibrino-hemorrhagic necrosis were observed in the ruminal sacs. Some of the necrotic masses were detaching and rolling up at the margins. Serosal adhesions to surrounding structures were observed over most lesions. A yellowish-white caseous mass with a blood clot on the surface was situated in the caudal fundus. The pyloric mucosa was studded with a few superficial erosions.

Day 33 (1). An estimated 500 ml of blood were found in the feed box utilized by this sheep 8 days prior to sacrifice. Several small necrotic and pale depapillated areas (2×1 , 2.5×2 , 3.5×1.5 , and 1×1 cm) were present in the ruminal sacs. The abomasal mucosa was hyperemic. The right lung was heavy and firm. Pericardial fluid was increased. The irradiated skin was firm and cyanotic. There were several areas of epilation. The exposed epidermis was dry, slate colored, encrusted and scaly. Desiccated serum clots were adherent to parts covered with wool.

Day 47 (1). An 11.5 cm Y-shaped dark-colored scar with a central erosion (2.5 x 0.5 cm) in the anterior ruminal sac and a pale depapillated area in the posterior ventral sac were present. The abomasal fundus was slightly edematous and hyperemic. There was a 4 x 2 cm area of necrosis in the caudal fundus and cephalic pylorus. The necrotic tissue was sloughing. Necrosis and sloughing involved 1.75 to 4 cm of three caudal fundic laminae. A 2 cm area of necrosis occurred between two laminae. The spleen was small and pale. The cardiac ventricles were dilated. The lungs were heavy, bluish in color and were studded with small yellow foci. A longitudinal band of epilation and superficial necrosis 20 mm in width occurred at the junction of the irradiated and nonirradiated skin.

Day 60 (1). The anterior ruminal sac had three stellate scars (3 x 2.5, 2.5 x 2 and 3.5 x 1 cm). Two scars had slit-like fistulous tracts which were walled off on the serous surface by scar tissue and/or adhesions. A 9 x 6 cm blue-gray scar straddled the posterior pillar. Two fistulous tracts which penetrated the ruminal wall were walled off by serosal adhesions. The surfaces of the tracts were covered with a yellowish-green exudate. A 9 x 6 diverticulum (6 cm in the caudal fundus and 3 cm in the cephalic pylorus) had developed. The mucosa of the diverticulum was necrotic. The abomasum in this area was adherent to the abdominal wall. Two 0.75 cm circular, elevated, pale fibrous plaques studded the cecal mucosa. Serous degeneration and atony involved the heart. The irradiated skin area was congested, and inelastic. A sharp line of demarcation occurred at the junction of the irradiated and nonirradiated region.

Histopathologic Observations. The parakeratotic layer of the rumen of control sheep varied from 2-4 to 6-8 cells in thickness. There were scattered intraepithelial microcysts. A few microabscesses were present in the epithelial layer. The rete pegs were of variable length. There were numerous focal areas of cell infiltration, principally lymphocytes, in the lamina propria. Several lymphoid follicles were observed in the abomasal lamina propria. An occasional superficial mucosal gland was dilated.

Only sheep exposed to GI irradiation are included in this phase of the report, since considerable data are already available on whole-body gamma irradiated animals and some are available on skin irradiation (Brown *et al.*, 1968; Bustad and McClellan, 1966).

Day 2 (1). Focal areas of swelling and "rounding up" of epithelial cells beneath the parakeratotic layer were occasionally observed. These foci were pale-staining and involved the entire epithelial thickness. Other foci of enlarged rounded cells in which the cytoplasm stained a dull red color were seen. The parakeratotic layer was absent over these areas. Foci of enlarged parakeratotic cells were occasionally observed. There were scattered single or multiple microcysts or intraepithelial cysts involving the stratum granulosum. There were focal areas of superficial necrosis of the abomasal mucosa which were not associated with vascular or cellular reactions. The lymphoid follicles appeared to be less cellular than those seen in control animals.

Day 3 (1). Focal areas of enlarged rounded cells in the parakeratotic layer and areas of "ballooning" (enlarged, rounded, pale staining cells) in the stratum granulosum were observed. A few foci of enlarged,

rounded, pale staining epithelial cells and small foci of edema of the omasal laminae were seen. There were a few small areas of superficial necrosis of the abomasal mucosa. Some superficial glands were dilated and a few small interstitial aggregates of polymorphonuclear (PMN) leukocytes were observed.

Day 4 (2). The parakeratotic layer was absent in areas of the rumen. Foci of enlarged, rounded, pale-staining epithelial cells with pyknotic nuclei were observed. The cytoplasm of some of these cells was stained a dull red color. Microcysts in the granulosum layer contained eosinophilic granular material. Small focal and 1 larger area of focal necrosis of the epithelium were observed. The change from normal to areas of necrotic epithelium was abrupt. There were foci of PMN, lymphocytes and mononuclear cells in the underlying lamina propria and aggregates of macrophages in the submucosa. An area of necrosis and sloughing of the epithelium of one omasal lamina was observed. There were several nidi of necrosis (superficial and deeper) of the abomasal mucosa. In some foci PMN infiltration had occurred. The submucosa was edematous.

Day 5 (1). In some papillae the parakeratotic layer was elevated forming a space which contained eosinophilic fluid and cellular debris. The detritus was the result of degeneration and necrosis of underlying epithelium. In some areas, the entire epithelial layer was destroyed. The underlying lamina propria was edematous and infiltrated with PMN cells. In other areas, the epithelium was focally enlarged, rounded and the cells separated by fluid. Several papillae were enlarged, the epithelium necrotic and the supporting lamina propria edematous and markedly

infiltrated with PMN cells. There were areas involving several papillae in which extensive and apparently rapidly developing plasma effusion into the lamina propria had markedly distended this supporting tissue. The plasma and fibrinous organization created a "honeycomb"-like effect. Necrotic epithelium was identified on the surface and sides of the masses. The submucosa was edematous. Focal areas of the omasal epithelium were enlarged, rounded, and pale staining. There were areas of frank epithelial necrosis. The underlying lamina propria was edematous and infiltrated with PMN cells. Small areas of necrosis, edema and PMN cell infiltration were observed in the abomasal mucosa. Some adjacent glands were dilated. A slight exudate of fibrin and cellular debris was adherent to the surface. The mucosa was edematous.

Day 7 (2). There were areas in which several necrotic papillae had sloughed. The congested, frayed condensed portion of the upper submucosa formed the ruminal lining. The epithelium of adjacent papillae was enlarged and rounded. There were numerous intraepithelial microcysts. Groups of papillae were distended with plasma and fibrin. There were large areas of fibrino-necrosis of the mucosa. Areas of hemorrhage and extremely numerous PMN cells were observed in the necrotic masses. The upper submucosa was edematous and extensively infiltrated with PMN cells. The blood vessels were dilated and the walls of some were necrotic. A lesser but appreciable inflammatory reaction affected the lower submucosa and extended into the underlying muscle layers. There was subserosal edema and inflammatory cell infiltration. Focal areas of epithelial necrosis and sloughing were seen in the omasum. There were nidi of enlarged, rounded epithelial cells with pyknotic nuclei. Small areas of

hemorrhages, edema and PMN infiltration were seen in the lamina propria. There were areas of hemorrhagic necrosis of the abomasal mucosa. In areas the necrotic-hemorrhagic mucosa appeared to have been "lifted up" by subsequent hemorrhage. A layer of necrotic epithelium covered the surface of underlying hemorrhage in some areas. Clumps or colonies of large rod-shaped bacteria were seen in the necrotic-hemorrhagic exudate. Glands in the mucosa adjacent to the hemorrhagic-necrotic area were dilated. Blood vessels above the muscularis mucosa were dilated, necrotic and thrombosed. The muscularis mucosa was interrupted in areas. The submucosa was edematous, hemorrhagic and markedly infiltrated with PMN cells. Some submucosal blood vessels were necrotic and thrombosed. Muscle bundles were separated by edema.

Day 11 (1). Large areas of fibrino-necrosis involved the mucosa of the rumen. The architecture was retained in most areas. In areas composed of several papillae, apparently extensive and rapid inflow of plasma into the lamina propria caused marked distention of the structures. Necrotic epithelium covered parts of the surface and sides of these areas. Epithelium of papillae adjacent to necrotic areas was either moderately normal or was enlarged and rounded. The submucosa underlying the large necrotic mucosal areas was necrotic and edematous. Cellular exudation was minimal. Numerous blood vessels were thrombosed and necrotic. In other areas the submucosa was edematous and hemorrhagic with extensive PMN infiltration. Muscle bundles were separated by edema and inflammatory cell infiltration. Beneath the severe mucosal and submucosal reactive areas, focal necrosis of muscle tissue occurred. There was an appreciable subserous edema. There were scattered nidi of

superficial necrosis and a large area of hemorrhagic necrosis of the abomasal mucosa. A large fibrino-hemorrhagic organization rested on the surface. Blood vessels at the base of the mucosa and upper submucosa were necrotic and thrombosed. The muscularis mucosa was penetrated in areas. The submucosa was markedly edematous and hemorrhagic. The muscle layers were stretched and thin.

Day 13 (1). There were large fibrino-necrotic masses involving the ruminal mucosa. In areas necrotic epithelium covered the surface of these necrotic masses. The parakeratotic layer was fairly well preserved. The epithelium of papillae at the border of the necrotic masses was enlarged and rounded. The nuclei were pyknotic. The supporting propria were markedly infiltrated with PMN cells. Some rete pegs were irregular in shape and of increased length. The submucosa was moderately edematous with areas of hemorrhage and was infiltrated with PMN, lymphocytes and mononuclear cells. Collagen fibers in the upper submucosa were enlarged, stained a dull red color, were anuclear and frayed. Some blood vessels were thrombosed and the walls necrotic. There was a large area of hemorrhagic necrosis of the abomasal mucosa. In an adjacent area the upper mucosa was necrotic and extensively infiltrated with PMN cells. In other areas a thin layer of necrotic mucosa covered an underlying thick layer of hemorrhage. Blood vessels beneath the hemorrhage at the base of the mucosa were necrotic and thrombosed. The muscularis mucosa was discontinuous in areas. The submucosa was markedly edematous and moderately hemorrhagic.

Day 15 (1). Large masses of fibrino-necrosis involved the ruminal mucosa. These masses were in areas covered by a parakeratotic layer.

In other areas the necrotic mucosa had apparently sloughed exposing a hemorrhagic-fibrous base. Epithelium of papillae adjacent to the necrotic tissue was enlarged and rounded. Rete pegs were thickened and of increased length. The lamina propria was infiltrated with PMN cells. The blood vessels were congested. The submucosa was edematous with hemorrhages and extensive PMN cell infiltration. Some blood vessels in the upper portion were necrotic. Muscle bundles were separated by edema and inflammatory cell infiltration. The subserosa was edematous. Focal areas of hydropic degeneration and superficial necrosis involved the abomasal mucosa. A moderate degree of interstitial edema was seen. There was a thin fibrino-cellular exudate on the surface.

Day 20 (1). In areas the ruminal mucosa had sloughed exposing a congested, necrotic condensed layer of the submucosa. Large areas of the mucosa were affected with fibrino necrosis. The surfaces were irregularly covered with parakeratotic epithelium. Perpendicularly directed thin bands of necrotic epithelium coursed through the necrotic masses. The submucosa was markedly edematous. Extensive PMN and moderate mononuclear cell infiltration was seen. There were scattered areas of hemorrhage and small aggregates of degenerated PMN cells. Several large vessels were dilated and empty, other vessels were necrotic, particularly in the upper portion of the submucosa. In one area a necrotic band of tissue extended from the edematous serosa through the muscle layers and submucosa to the necrotic mucosa. Scattered nidi of superficial necrosis of the abomasal mucosa were seen. These were not associated with a cellular or vascular reaction. Some glands were dilated. There were scattered foci of PMN cell infiltration. The submucosa was

markedly edematous and some blood vessels were dilated and engorged.

Day 59 (1). The lining of large areas of the rumen consisted of a mixture of granulation tissue and fibroblasts. The fibroblasts were oriented parallel to the surface which was necrotic and "ragged". Underlying collagen fibers were swollen and capillaries were numerous. In other areas the epithelium was dark staining and 2 to 3 cells in thickness. The nuclei were oriented parallel to the surface. There were no papillae and the surface was undulating. The epithelium appeared to be poorly attached to the underlying base. Rete pegs were either absent, or short, or a few were long and irregular. In other areas the surface consisted of minute, relatively normal appearing lamina propria pegs which lacked an epithelial covering. There was no vascular or cellular response in the underlying tissue. The submucosa was edematous and extensively infiltrated with macrophages, many containing ingested foreign material. There were areas of necrosis in the muscle layers. The walls of some arterioles were eccentrically thickened. Changes in the abomasal mucosa varied from: dilated glands; atrophy; interstitial edema, atrophy and glandular degeneration with moderate mononuclear, fewer lymphocytes and PMN cell infiltration; focal superficial necrosis; necrosis of the entire mucosa; to ulcer formation. A large area of the submucosa which formed the base of the ulcer was replaced by vascular granulation tissue and fibroblasts. This tissue was moderately infiltrated with macrophages and PMN cells. A few colonies or clumps of large gram-positive bacterial rods were seen beneath the necrotic surface. Coagulation necrosis involved another large area of the submucosa forming the base of the ulcer.

A number of dilated, necrotic, thrombosed blood vessels were seen. Nuclear debris and a few macrophages were scattered in this necrotic tissue. A band of caseation necrosis involved the lower submucosa and part of the thin muscle layer. The atrophic muscle tissue was infiltrated with inflammatory cells. The serosa could not be identified. The muscle tissue rested on a rather thick layer of collagenous fibers. Atrophic muscle bundles containing islands of granulation tissue and fat tissue formed the external surface.

⁹⁰Y Excretion. Fecal ⁹⁰Y excretion levels, as a percentage of the total dose, increased rapidly and reached a peak by the third or fourth day (Fig. 1). Fecal radioactivity declined rapidly with discontinuation of feeding of the fallout simulant with a half-time of less than one day. Ninety-nine percent of the ⁹⁰Y had been excreted or decayed by 8 to 10 days after dosing. There were no significant differences in excretion among the various treatment groups.

Feed Consumption and Weight Changes. The feed consumption was measured for 66 days and the only feed refusals were by those sheep fed ⁹⁰Y-labeled sand alone or in combination with the other treatments (Fig. 2). These feed refusals started 3 to 5 days after initiation of treatment, became severe by 10 to 15 days, and most of these treated sheep returned near to the control levels by the end of the 66 days.

Weight losses of 20% of body weight in 18 days by those fed ⁹⁰Y sand reflected the reduction of feed intake (Fig. 3). However, weight losses averaging about 14% by those skin irradiated alone and in combination with WB irradiation were observed by day 50, even though these sheep were consuming the same amount of feed for the first 38 days of

this period. The weight loss by these skin-irradiated sheep was a gradual decline and did not show the abrupt changes of those fed ^{90}Y sand. The rapid increase in weight at 10 weeks is a reflection of ad libitum feeding of all sheep after 8 weeks.

Zinc. In vitro uptake of ^{65}Zn by erythrocytes from controls, ^{90}Y fed and control pair-fed to the level of feed intake by the ^{90}Y -fed sheep is shown in Fig. 4. These data show that restricted feed intake increases the in vitro uptake of ^{65}Zn but not nearly to the extent of those fed ^{90}Y sand. Skin and WB irradiation did not significantly affect in vitro uptake during the 42-day period, but skin irradiation significantly increased the ^{65}Zn uptake after a period of 8 weeks as shown in Fig. 5. By 32 weeks, in vitro ^{65}Zn erythrocyte uptake increased to 8% comparable to the peak at 4 weeks in GI irradiated sheep. This level of twice normal has now persisted for 50 weeks in some skin-irradiated sheep.

Stable Zn in plasma was only slightly affected as shown in Fig. 6. Also the procedure for ^{65}Zn uptake was specific for this ion since no differences were found due to treatment when ^{75}Se and ^{54}Mn were incubated under the same procedures (Figs. 7 and 8).

Plasma Iron. The plasma iron concentration as a function of time for the control group and skin and GI irradiated groups is shown in Fig. 9. Plasma iron concentration in the control sheep decreased slightly during the second and third weeks after the studies began, probably due to the considerable amount of blood being withdrawn from these animals. The pattern of the sheep fed radioactive sand was not essentially different from that of controls. Plasma iron concentrations

of the sheep which received whole-body irradiation are not shown but were also similar to that of the controls. There was, however, a marked decrease in the plasma iron concentration in the skin-irradiated sheep. This decrease was evident by the second week, and showed some evidence of recovery at the fourth week after treatment. Plasma iron concentration in the sheep receiving combination treatments is not shown in the graph, but all sheep which received skin irradiation, regardless of other treatments, showed the decrease in plasma iron during the second and third weeks after treatment, with recovery becoming evident by the fourth week. This same type of phenomena has been observed in dogs with turpentine-induced abscesses, and was possibly due to an inhibition of the release of iron from the reticuloendothelial system.

⁵⁹Fe Clearance and Incorporation into Erythrocytes. Plasma iron clearance and percent of iron incorporated into new erythrocytes prior to treatment and 4 weeks after treatment are shown in Fig. 10. Again it can be seen that skin-irradiated animals showed greater changes after treatment in both rate of iron clearance and percent of iron incorporated into new red cells. These data are also consistent with an inhibition of release of iron from the reticuloendothelial system.

Plasma Copper Concentration. Copper concentration in the plasma as a function of time after treatment for the control group, the whole-body irradiated group and the groups which received beta irradiation of the skin and GI tract is shown in Fig. 11. The middle graph shows the whole-body irradiated group compared with the control group. It can be seen that there was essentially no difference between the two groups, with a possible exception of values obtained at 10 weeks after treatment.

At the present time, there are insufficient data to determine whether or not plasma copper concentration in the whole-body irradiated group did increase at this time. In the GI irradiated group there was a marked increase in plasma copper concentration at 2 weeks after treatment and then a decrease to near control levels, with again a suggestion of a gradual increase in copper concentration beginning at about 10 weeks after treatment. The skin irradiated group showed increased plasma copper concentration at 3 weeks after treatment, a decrease at approximately 5 weeks after treatment, which may or may not have been significant, and then a subsequent gradual increase. In these Figures, standard errors are shown at selected points.

It can be seen that whole-body irradiation combined with any other type of radiation injury produced a greater change in plasma copper concentration, even though whole-body irradiation alone had no effect on plasma copper levels.

Plasma Mg and Ca. GI, GI + WB, and GI + Wb + Skin treatments reduced plasma Mg levels while the other treatments had little effect (Fig. 14). During the period of observation, the combined treatments most severely affected plasma Mg, but neither Skin nor WB alone affected this parameter. No hypomagnesemia tetany was observed in these sheep, but the low plasma levels observed in combined treatments could have contributed to the anorexia shown in Fig. 1 since these plasma levels were similar to those reported by McAleese, et al. (1961) for hypomagnesemic sheep.

Plasma Ca levels of all sheep fed the ⁸⁰Y-labeled sand were drastically reduced, particularly at 3 weeks postirradiation (Fig. 13).

This decrease may merely be a reflection of the decreased feed intake and thus Ca intake during this period, but no pair-fed studies were conducted. A slight decrease was also observed in plasma of sheep receiving whole-body and skin irradiation.

Dosimetry. The range and average dose to various points in the GI tract are shown in Table 2. The first two areas of the rumen were those in which lesions were consistently found at necropsy. The doses measured in the rumen may be too low due to the fact that the small sand particles could lodge in areas at the base of the papillae which were inaccessible to the encapsulated dosimeters. In the rumen and abomasum, variability within the same animal was as great as the variability among animals. It was not uncommon to recover dosimeters on the same string, less than 5 cm apart, which indicated doses differing by a factor of 4.

B. Beef Cattle

During the reporting period four replications of the treatments listed in Table 1 were initiated and the results are similar to those obtained with the sheep. The combined effects of the three treatments on mortality and the gross pathological observations in the necropsies indicate that the cattle were more severely affected than the sheep. Not only was the rumen and abomasum more severely affected, but more lesions were found in the reticulum, omasum, and intestines. The damage to the abomasum was still the most significant damage in cattle. During the reporting period all 3 steers given the three types of radiation died in 14 to 28 days and one of the 3 on a combined WB and GI died during this time. The reporting period covered only 9 days for the fourth replication. None of those on the other treatments died.

Complete results on beef cattle will be reported when the dosimetry and other replications are completed and the data are analyzed.

DISCUSSION

General. These data clearly demonstrate that grazing sheep and cattle exposed to radioactive fallout under these simulated conditions were severely affected by ingested fallout and skin irradiation at levels where whole-body gamma irradiation alone had minor effects. The high mortality rate and the drastic weight losses would severely affect livestock productivity. Anorexia and weight loss would severely affect milk of dairy cattle for the duration of the current lactation and meat production of meat-producing ruminants for many months. The weight loss from skin irradiation would be expected to be much greater for animals remaining outside during the winter months than in our sheep kept under a shed partially enclosed on three sides. In the summer months, the fly problem required vigilance to prevent severe damage from fly larvae. Under livestock range conditions, it is unlikely that this type of care could be taken to prevent animal losses from secondary effects.

If there were conditions of food shortages, those animals surviving but severely affected could still be used for food even if they were developing hernias, fistulae and/or severe dorsal skin injury.

Fallout Assumptions. In order to assess the vulnerability of livestock to fallout radiation, it is necessary to determine the likelihood of sheep and cattle being exposed to the levels of GI, WR and Skin irradiation used in these studies. From the data available, it appears that sheep and cattle grazing downwind from a surface burst of at least a megaton-size weapon could be exposed to these levels.

If we assume that the 680 g of alfalfa consumed daily by the sheep is equivalent to the forage grazed from a pasture area of 6.8 m^2 , then using the NAS-NRC (1963) assumption of H + I dose rate in which 100 R/hr is produced from 6100 mCi/m^2 which decays to 161 mCi/m^2 at 24 hr, then the consumption of $\approx 72 \text{ mCi}$ on day 1 would represent about 7% retention of fallout on the forage for sheep. With an average of 10 mCi/g of sand, the 161 mCi/m^2 would give 16 g of fallout/ m^2 on the pasture land.

The steer calves (9 to 12 months of age) consumed 2.7 kg of alfalfa daily which would be equivalent to the average forage consumed on 28 m^2 of pasture. Using the same procedure as used for sheep, then the calculated 24-hr value would represent about 9% retention on the pasture forage. With some concern for all the variables an estimate of 7 to 9% retention on forage would probably be a safe assumption for the first day for average sheep and cattle pastures in an area where the whole-body gamma irradiation amounted to about 240 R in the first 72 hr. Research in progress at Colorado State University indicates that this level of retention is realistic using this same size sand under their grazing conditions (J. E. Johnson, personal communication). The fallout arrival time would affect the total radioactivity consumed not only from radioactive decay but also from the grazing habits of the animals and the quality of the forage. Sheep and cattle normally graze in early morning and late afternoon with sheep grazing more closely than cattle. Also more grazing time is required and more soil is ingested on over-grazed pastures.

The rectangular skin irradiation sources ($28 \times 43 \text{ cm}$) covered about 12% of the body surface of sheep (Brady, 1945). In using two of these

sources for each steer, the exposed area amounted to about 8% of the body surface. Originally, it had been planned to use three sources for each steer to also cover 12% of the body surface, but this was not feasible under the conditions of this study.

Pathology. Ingested ^{90}Y -labeled sand particles appeared to collect in rather definite areas in the ruminal compartments. Rhythmic contractions of the musculature and compartmentalization produced by the pillars probably influenced or determined the areas of sedimentation. The ruminal sac was frequently the site of the most extensive involvement. Some particles appeared to be more or less permanently confined to these areas by lodging between papillae. Unfortunately, numerous sand particles were still present when the tissues were processed for microscopic examination. Effusions of plasma and subsequent fibrin formation also may have been a factor in trapping the radioactive material in these areas.

The parakeratotic layer appeared to be relatively resistant to radiation injury. Necrosis of the mucosa and luminal portion of the submucosa in more extensively affected areas, concomitant or subsequent rapid and excessive outflow of plasma, fibrin formation and extensive PMN infiltration followed and participated in the development of the characteristic, albeit not pathognomonic, lesion. The necrotic mass was rather friable and often not too firmly anchored to the underlying tissue. With time and healing of the underlying tissue the necrotic mass was progressively detached and ultimately a variable-sized scar remained. How completely and efficiently healing occurs is not too clear at this time. Scars bearing superficial erosions and/or ulcers,

exudation and adherent necrotic tissue have been repeatedly observed up to 12 months after treatment. The bulky fibrous roughage-type ration was probably more irritating and retarded healing to a greater degree than would have the pelleted alfalfa.

If the quantity of radioactive material was appreciable focally, the inflammatory-necrotizing lesion was progressive and involved the entire thickness of the ruminal wall resulting in adhesions to adjacent structures. It is assumed that the ruminal fistula developed as the result of a concentrated local collection of radioactive material, rapidly developing adhesions, and progression of the lesion. There is no evidence that microorganisms were concerned in the pathogenesis of the lesion. Adhesions in apparently clinically recovered animals could be expected to interfere with rumen function and efficiency in proportion to the number and extent of adhesions. Intestinal strangulation could possibly occur as the result of such adhesions.

Only relatively minor lesions have been observed in the reticulum and omasum. The small yellow sessile-based nodular elevations infrequently seen on the reticular mucosa lead one to suspect that they are composed primarily of fibrin and necrotic tissue and that the mechanism of their development is similar to that of the ruminal lesions. This has not been confirmed by microscopic examination. Small superficial linear erosions, small hematomata and in one instance, a yellowish friable nodule adherent to the ventral free surface of two major laminae constitute the omasal changes. In consideration of the extent of ruminal and abomasal changes, more extensive reticular and omasal lesions were expected.

Edema, hyperemia and large local areas of hemorrhagic necrosis were rather consistently present in the abomasum. Edema and hyperemia were generalized. The caudal fundic area was the principal site of the severe hemorrhagic-necrotic lesions. The cephalic pylorus was appreciably less involved. Hemorrhagic participation in abomasal lesions was a conspicuous feature. In many cases, the reaction involved the entire thickness of the wall of the organ leading to adhesions to adjacent structures. In severe cases portions of the organ prolapsed through a hernial ring in the ventral abdominal wall. Possibly a sequence of closely related events were responsible for the prolapse; i.e., heavy local concentration of radioactive material, severe and rapidly developing inflammatory-necrotic lesions involving all coats of the abomasal wall and adhesions to the abdominal wall.

The absence of significant intestinal changes was not anticipated. Congestion, edema, and in a few cases focal necrosis of the duodenal mucosa have been seen. Two small circular fibrotic elevations were found in the cecal mucosa of one sheep. Since parasitic-induced nodules have been in evidence in the intestinal wall of most sheep, such lesions could have been the result of parasitism.

Large stellate scars are frequently observed in the rumen of well-conditioned cattle at slaughter. Usually, these animals have shown no signs of anorexia or illness. The scars are reported to be the sequela of acute rumenitis and conceivably could be anatomically comparable to the lesions in sheep with ^{90}Y -induced rumenitis. The abomasal lesions were regarded as the principal cause of anorexia, illness, and death in the sheep although the ruminal changes undoubtedly were contributory.

Examination of a limited number of animals exposed to combined types of irradiation indicates a synergistic effect as reflected by increased severity of lesions and reduced developmental time. The causes of respiratory and circulatory involvement in skin-plaque irradiated animals was not apparent.

Blood Studies. WB gamma-irradiated animals developed the expected drop in WBC and platelet counts but these effects were not observed in the other single treatments since the hematopoietic system was sufficiently protected from the beta irradiation. The stimulation of platelets by GI and Skin irradiation is similar to data obtained by giving subcutaneous irritants to animals.

The data on in vitro erythrocytic uptake of ^{65}Zn indicate that these beta-irradiated animals were zinc deficient (Berry et al., 1965). However, plasma zinc levels were only slightly reduced and no effort was made to restrict the dietary zinc intake. The 17 ppm of zinc in the alfalfa pellets, the 100 ppm in the tracemineralized salt, and the water containing up to 2.5 ppm of zinc should have been sufficient to prevent a zinc deficiency.

Uptake of ^{75}Se and ^{54}Mn was unaffected by irradiation treatment, therefore the ^{65}Zn increased uptake was probably not a membrane permeability effect. The erythrocytes from some of the skin-irradiated sheep taken as long as 12 months after exposure still showed about twice as much uptake as the controls. These data demonstrate that this effect is not from irradiation of the erythrocytes since it has been shown that sheep erythrocytic life span is 157 days (Wright, 1965). Since zinc is a component of many enzyme systems and is involved in wound healing

(Strain and Pories, 1969; Husain, 1969), it is postulated that the in vitro uptake of ^{65}Zn is an indication of the extra need for zinc in healing of the GI and Skin wounds from beta irradiation. Most of the GI repair occurred within 60 days, while most of the skin areas have not healed in 12 months, which corresponds with the times of elevated ^{65}Zn uptake. The delay in the ^{65}Zn uptake by erythrocytes from the skin-irradiated sheep is probably due to the lack of any healing during the first 50 days.

The decrease in plasma Fe and the increases in ^{59}Fe clearance rates from plasma and incorporation into the RBC in the skin-irradiated sheep could conceivably be explained by an inhibition of iron release from the reticuloendothelial system, but more evidence is needed to test this theory. Elevated serum Cu levels have been detected in a variety of pathological conditions including acute and chronic infection. However, the mechanism by which Cu is involved is poorly understood. The synergistic effect of whole-body gamma irradiation on plasma Cu levels of GI and Skin-irradiated animals is not understood. The depressed plasma levels of Ca and Mg levels in the GI-treated sheep is probably most accurately explained by the anorexia and thus a marked decrease in dietary Ca and Mg.

Dosimetry. The dosimetry data shown in Table 2 is an indication of the tremendous variability found in a uniform group of sheep fed identical rations containing the same quantity of ^{90}Y -labeled sand. It is very evident that due to "pocketing" of the sand that doses varied by a factor of 10 within the same site. These data can only be used to give a rough estimate of the radiation during the 8- to 10-day period.

Also it appears that biological response is much more reliable here than dosimetry data.

Beef Cattle. Very preliminary data on the cattle indicate that sheep were good models for cattle but that there are differences. Mortality appears to be higher on the three combined treatments with cattle and lower on the single and dual treatments than in sheep. Also the GI effects appear to be more severe in the cattle than sheep. However, a greater total activity and a different activity per kilogram of body weight is being used in the study. Additional numbers and GI dosimetry data on cattle will be required before definite conclusions can be drawn.

Predicted Effects on Livestock Production. The data reported in this paper clearly show that simulated fallout irradiation affects sheep and cattle more severely than only sublethal doses of whole-body gamma irradiation. Not only does it reduce the LD_{50/60} by at least 30%, but it also would severely affect productivity and usefulness of the survivors. Data in Table 3 show the previous estimates on LD₅₀ considering gamma irradiation only in comparison with the present data considering GI, Skin, and WB combined. No data are available on swine and poultry. The combined effects might be a problem in swine, since many are maintained on pastures, but poultry production practices are such that no problem is expected.

Note: The research was conducted according to the principles enunciated in the "Guide for Laboratory Animal Facilities and Care" prepared by the National Academy of Sciences-National Research Council.

SUMMARY

Sheep fed ⁹⁰Y-labeled fallout simulant at the rate of 2.5 mCi/kg to simulate 7% forage retention developed anorexia, diarrhea, and weight loss. Feed intake of the survivors usually returned to normal within 60 days. Beta irradiation of 57,000 rads to the dorsum of sheep, equivalent to 12% of the body surface, severely affected the skin and reduced body weight by 15% with no reduction in feed intake. Whole-body gamma irradiation of 240 R at 1 R/min affected only the platelets and white blood cells with no mortalities. These three types of irradiation given singly had little effect on mortality but when the three were combined, 4 of 8 sheep died within 60 days. In addition to mortality losses the loss of weight would severely affect the productivity of animals exposed to these levels of fallout. Preliminary data on cattle indicate that the combined radiation insults would be more detrimental to cattle than to sheep.

Mineral changes in blood included increased in vitro ⁶⁵Zn erythrocytic uptake and increased plasma copper in blood from GI and Skin irradiated sheep. Plasma calcium and magnesium were decreased in GI irradiated sheep while plasma iron was decreased from skin irradiation.

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Table 1. Sheep Mortality from Gastrointestinal, Whole-body, and Skin Irradiation

Treatment	No. animals	No. of deaths		Total
		61 days	Total	
Control	8	0	0	
WB (240 R gamma)	8	0	1*	
Skin (57,000 rads beta)	8	1*	3	
GI (2.4 mCi ^{90}Y /kg)	8	1	3**	
GI + Skin	8	0	1**	
WB + Skin	8	0	2	
GI + WB	8	3	4**	
GI + WB + Skin	8	4	4	

* Accidental deaths.

** Four animals were sacrificed following the development of ruminal and abomasal fistulae.

Table 2. Absorbed Dose to Various Points in the Gastrointestinal Tract of Sheep Fed 2.5 mCi ^{90}Y Sand/kg

Location	No. of animals	Absorbed dose	
		Range (kilorad)	Mean (kilorad)
Rumen			
Ventral sac	4	0.5 - 5.3	2.3
Caudodorsal blind sac	2	0.5 - 4.9	1.8
Dorsal sac	2	0.3 - 0.8	0.6
Abomasum			
Fundic region	3	4.8 - 35	12.8
Pyloric region	4	1.0 - 10.2	3.5
Reticulum	3	0.8 - 2.6	1.5
Omasum	4	1.1 - 5.5	3.3
Intestine	11	0.1 - 4.2	1.7

Table 3. Estimated 50% Lethal Dose to Food Producing Animals from Nuclear Fallout Radiation (Röentgens of gamma)

	LD _{50/30} NRC (1963)	Page (1968)	LD _{50/60} Present data*
Cattle	550	495	< 240
Sheep	550	340	~ 240
Swine	550	675	?
Poultry	800	---	?

* The present data are based on grazing animals with a combination of skin plus ingested fallout from 7 to 9% forage retention.

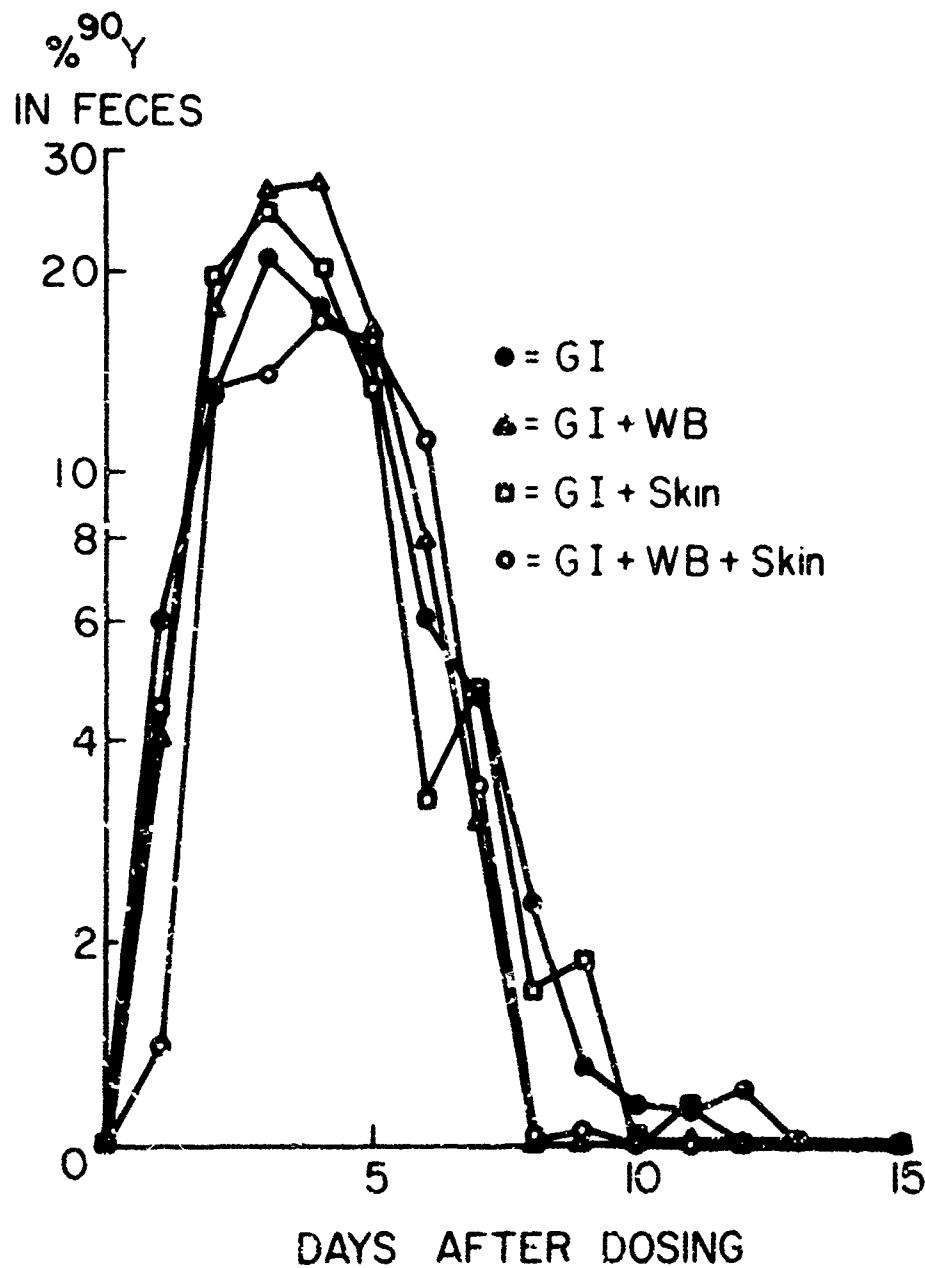


Figure 1. Fecal Excretion of ⁹⁰Y from Sheep Fed 2.4 mCi/kg for 3 Consecutive Days.

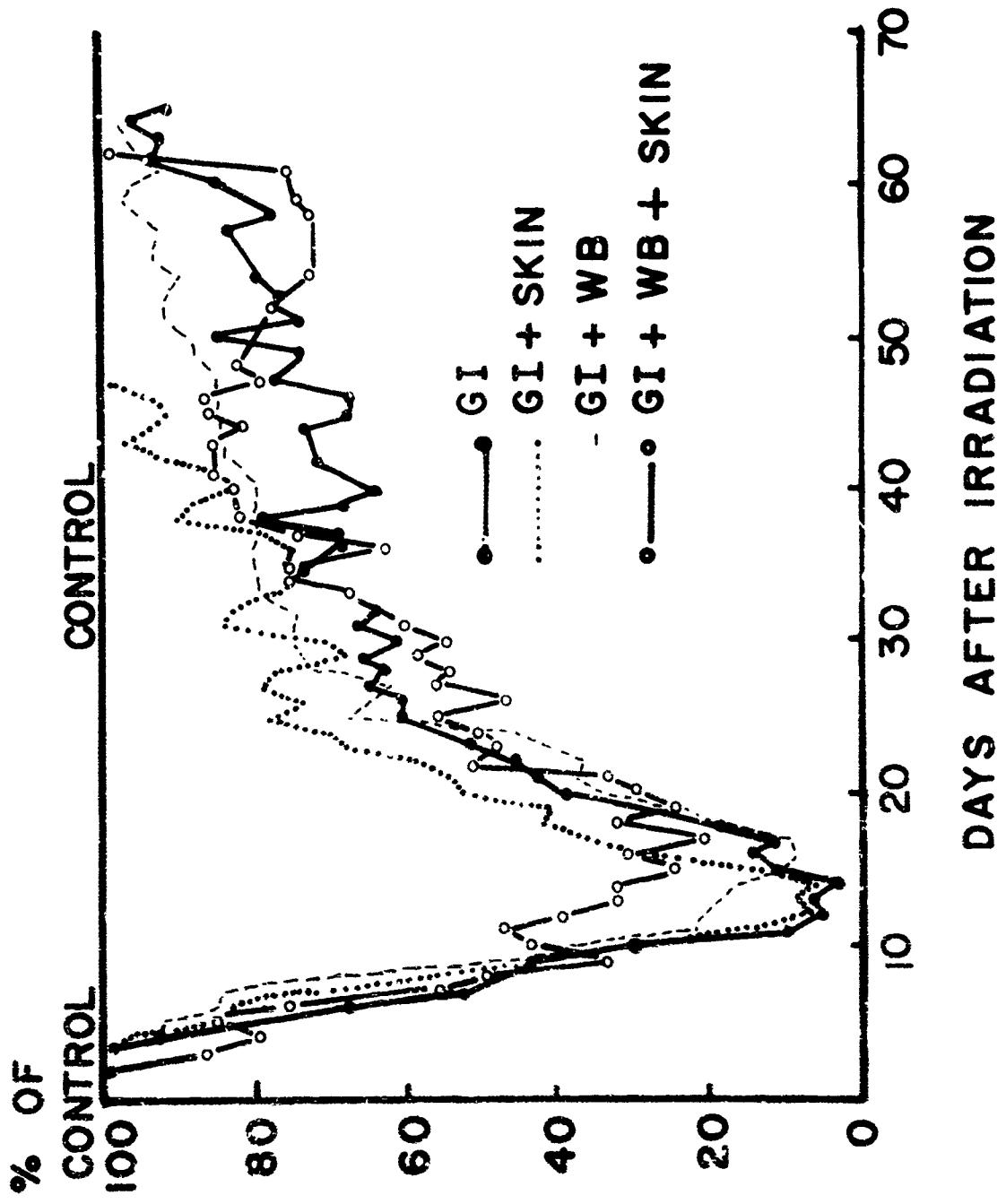


Figure 2. Radiation Effects on Feed Intake by Sheep. Intake of WB, Skin, and the Combined WB and Skin Was Identical with the Controls.

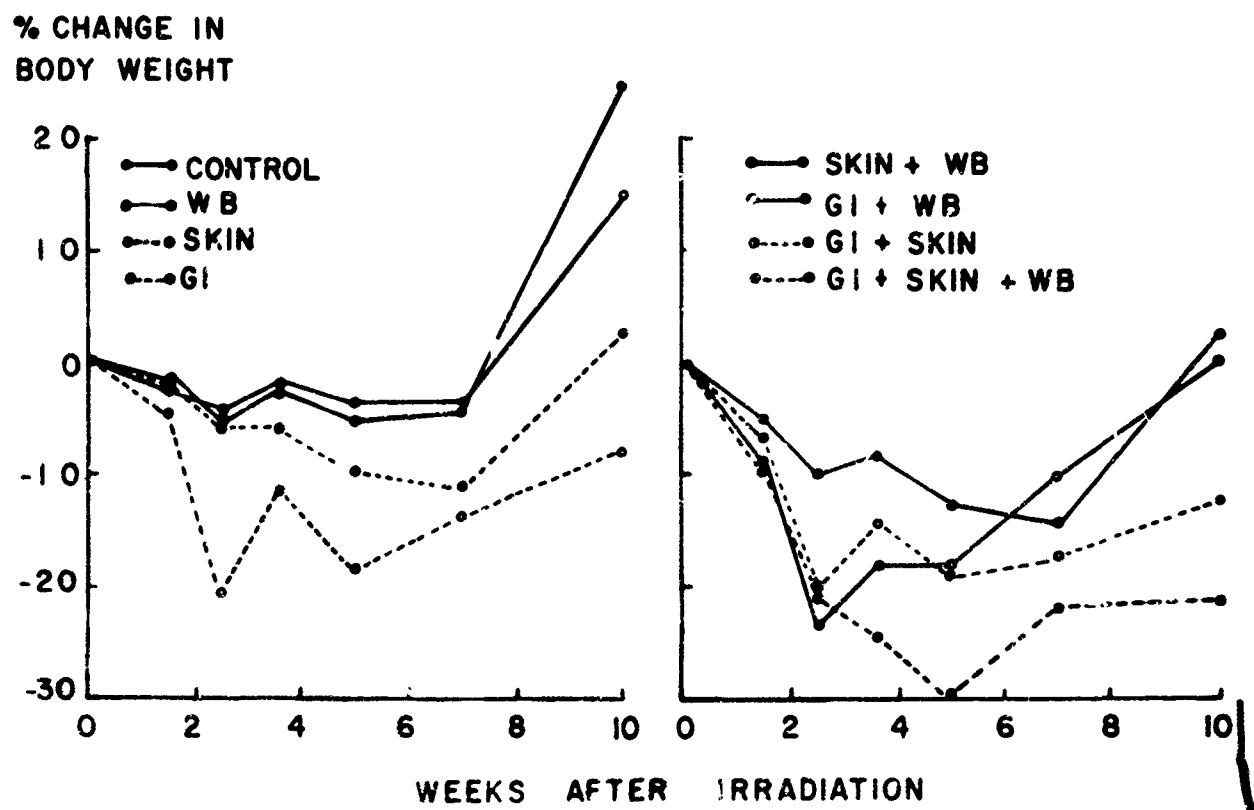


Figure 5. Radiation Effects on Body Weight of Sheep. Feed Intake Was Restricted to Maintenance Level for 7 Weeks.

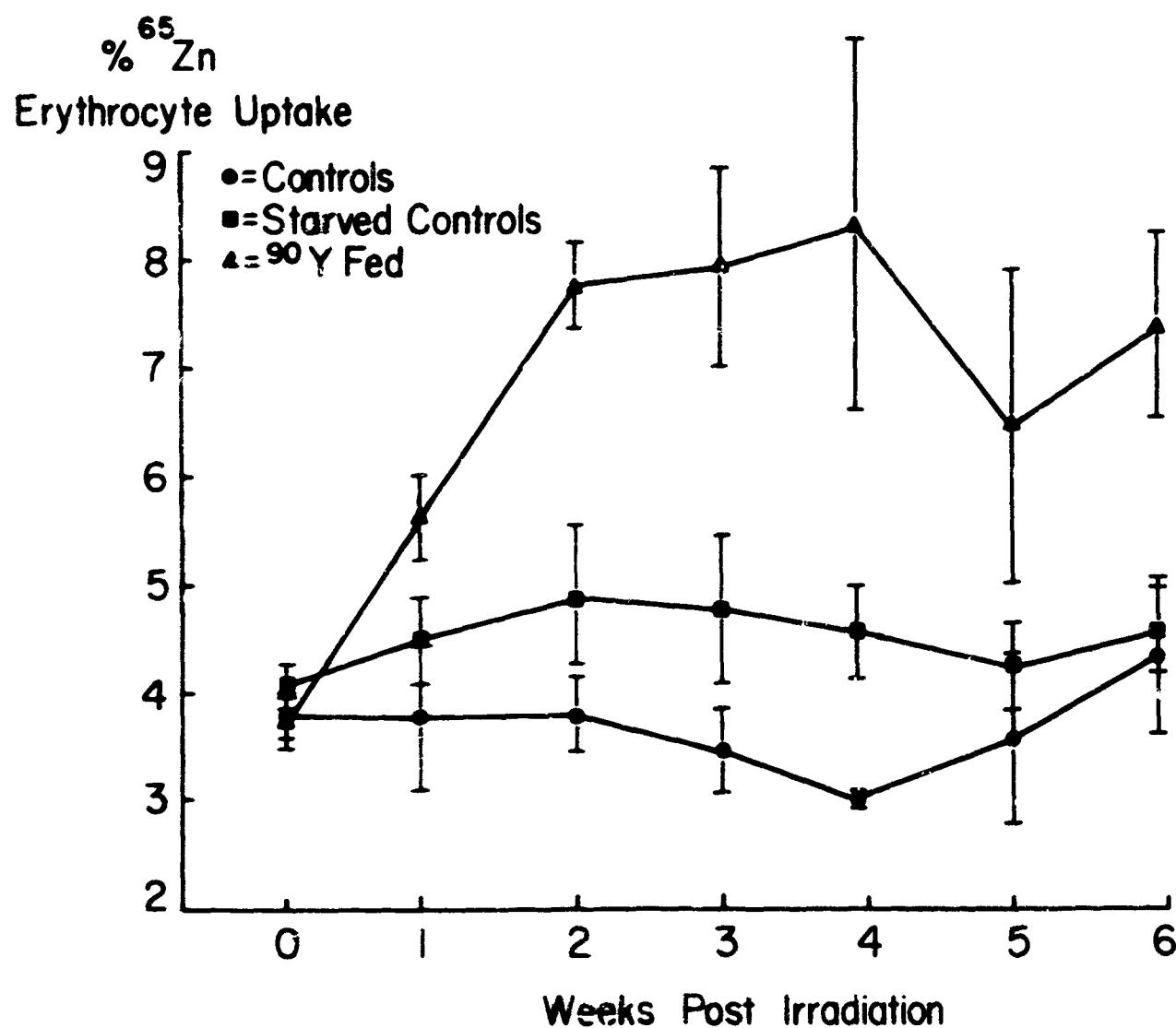


Figure 4. Effect of ^{90}Y Feeding and Starvation on In Vitro Uptake of ^{65}Zn by Sheep Erythrocytes. The Starved Controls Were Restricted to the Feed Intake of Those Fed ^{90}Y .

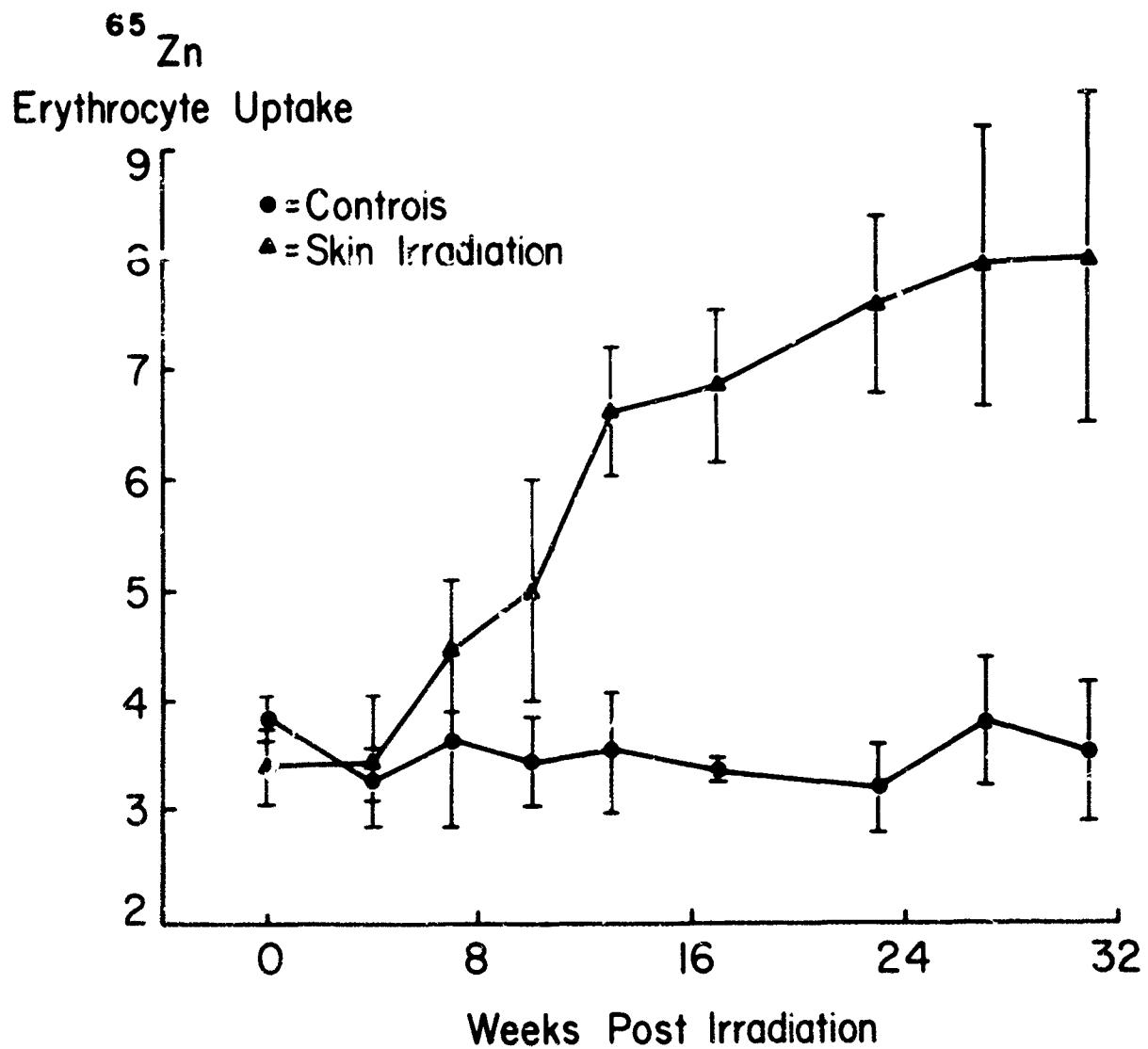


Figure 5. Effects of Skin Irradiation on In Vitro ^{65}Zn Uptake by Sheep Erythrocytes.

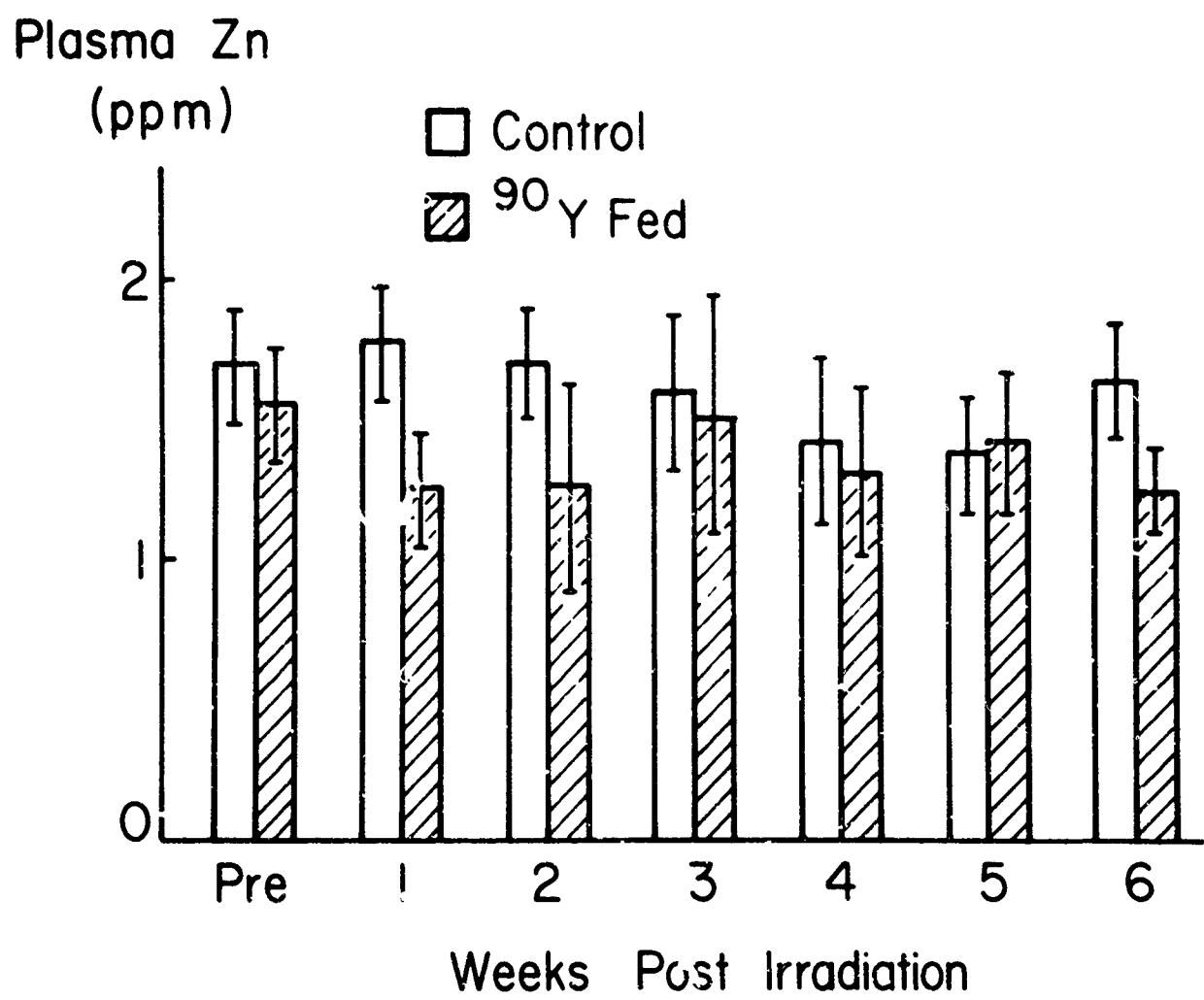


Figure 6. Plasma Stable Zn in Sheep Fed ^{90}Y -Labeled Sand and in Control Sheep.

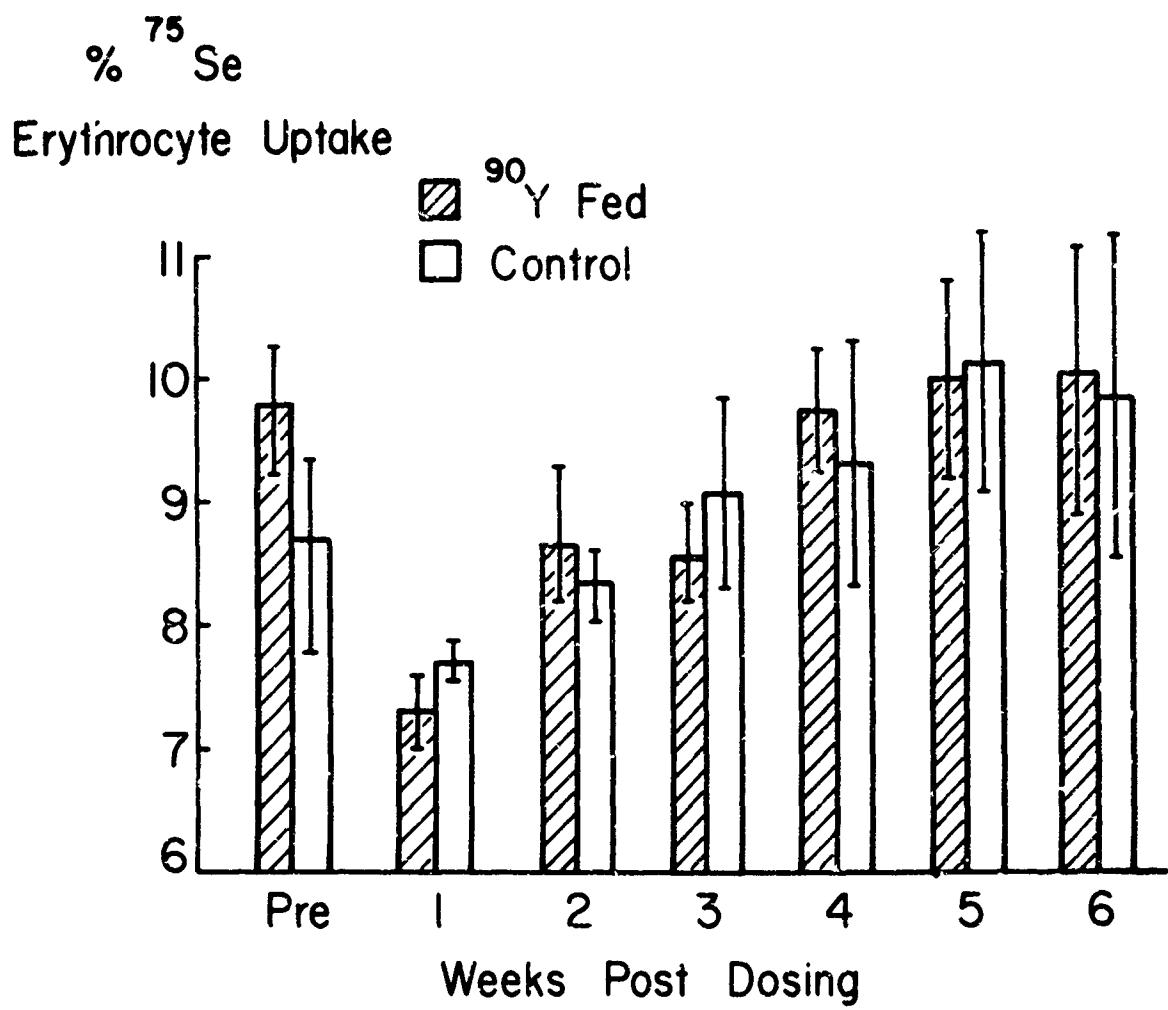


Figure 7. Effects of ^{90}Y Feeding on In Vitro Uptake of ^{75}Se by Sheep Erythrocytes.

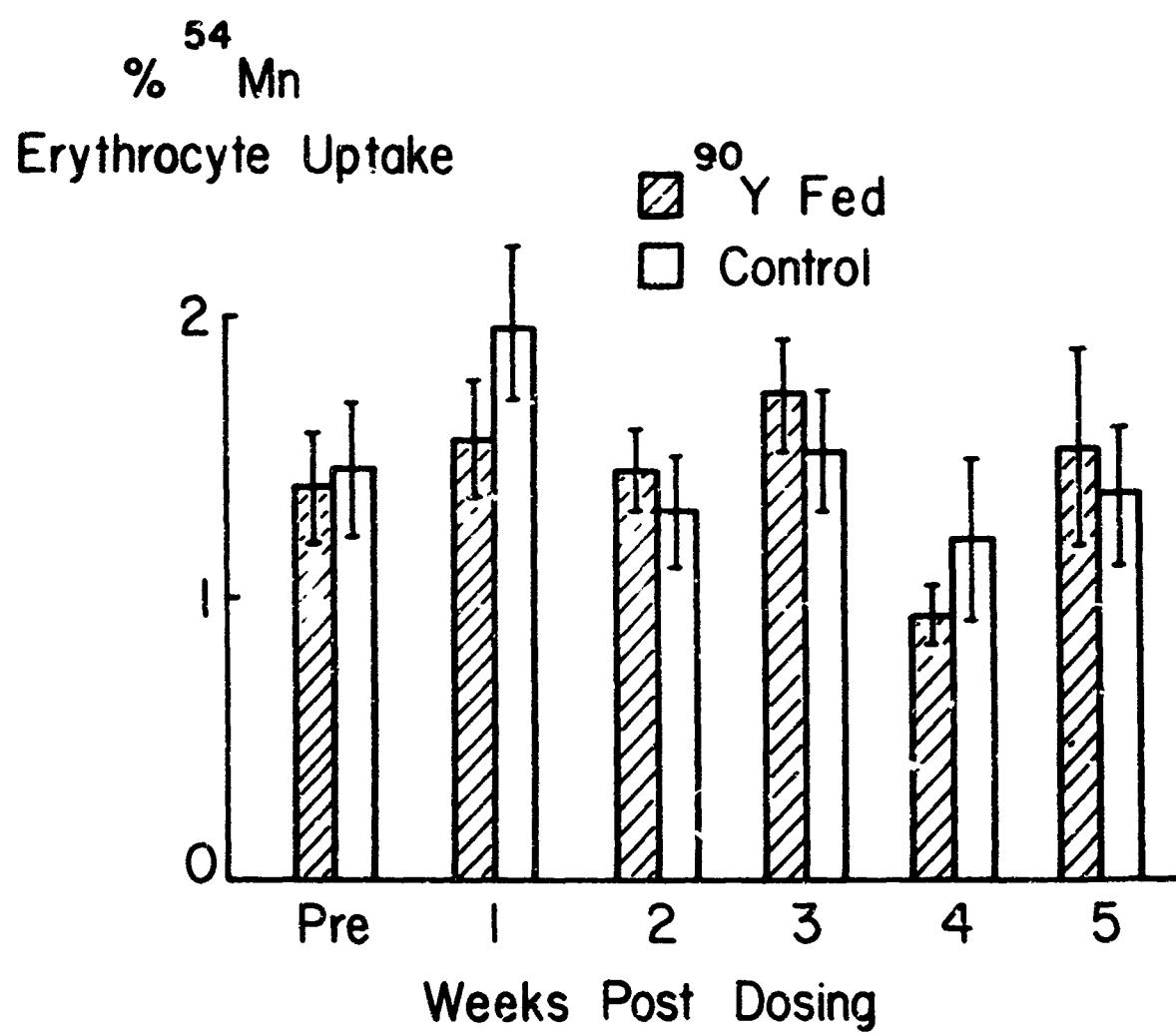


Figure 8. Effects of Feeding ^{90}Y on In Vitro ^{54}Mn Uptake by Sheep Erythrocytes.

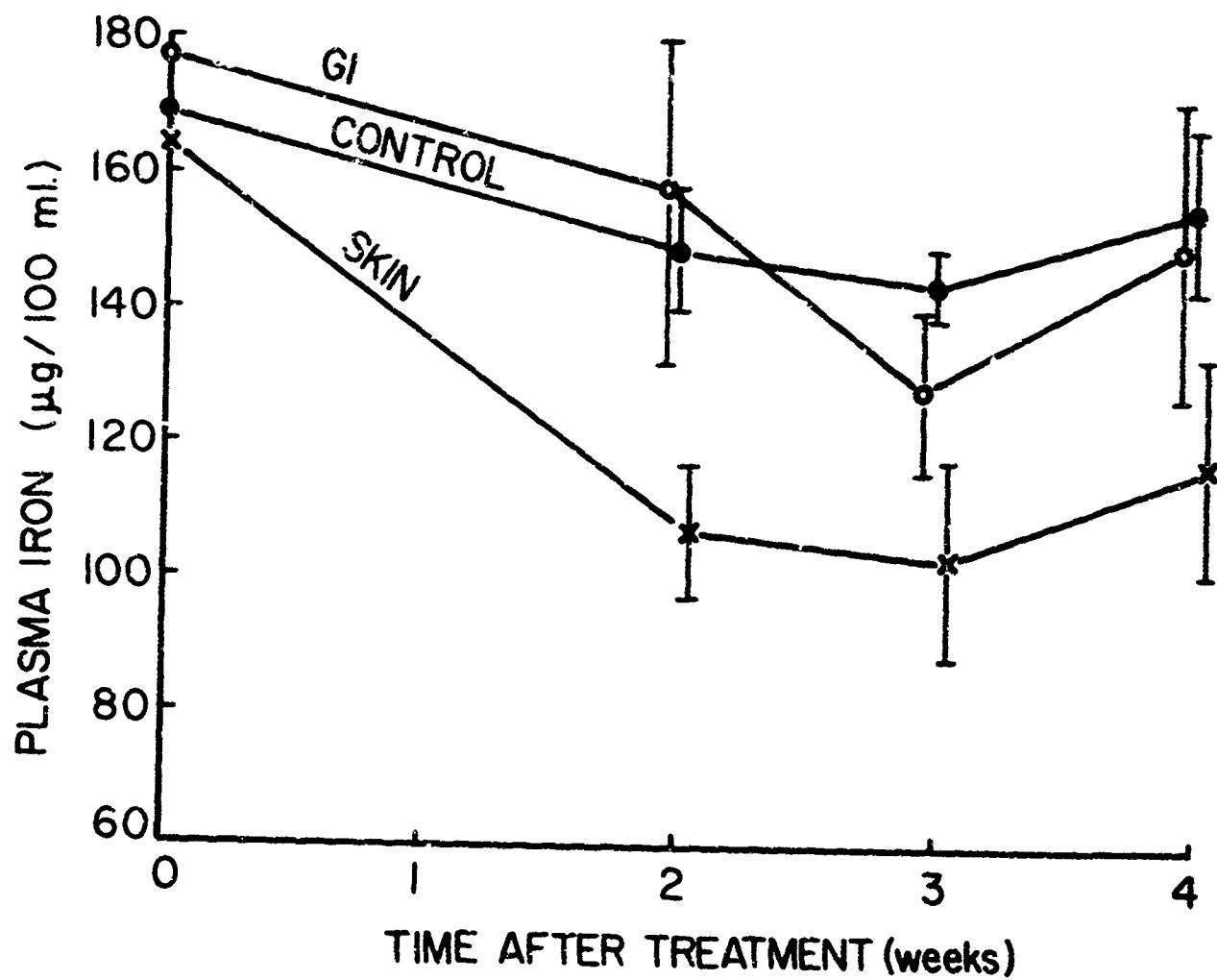


Figure 9. Radiation Effects on Plasma Fe in Sheep.

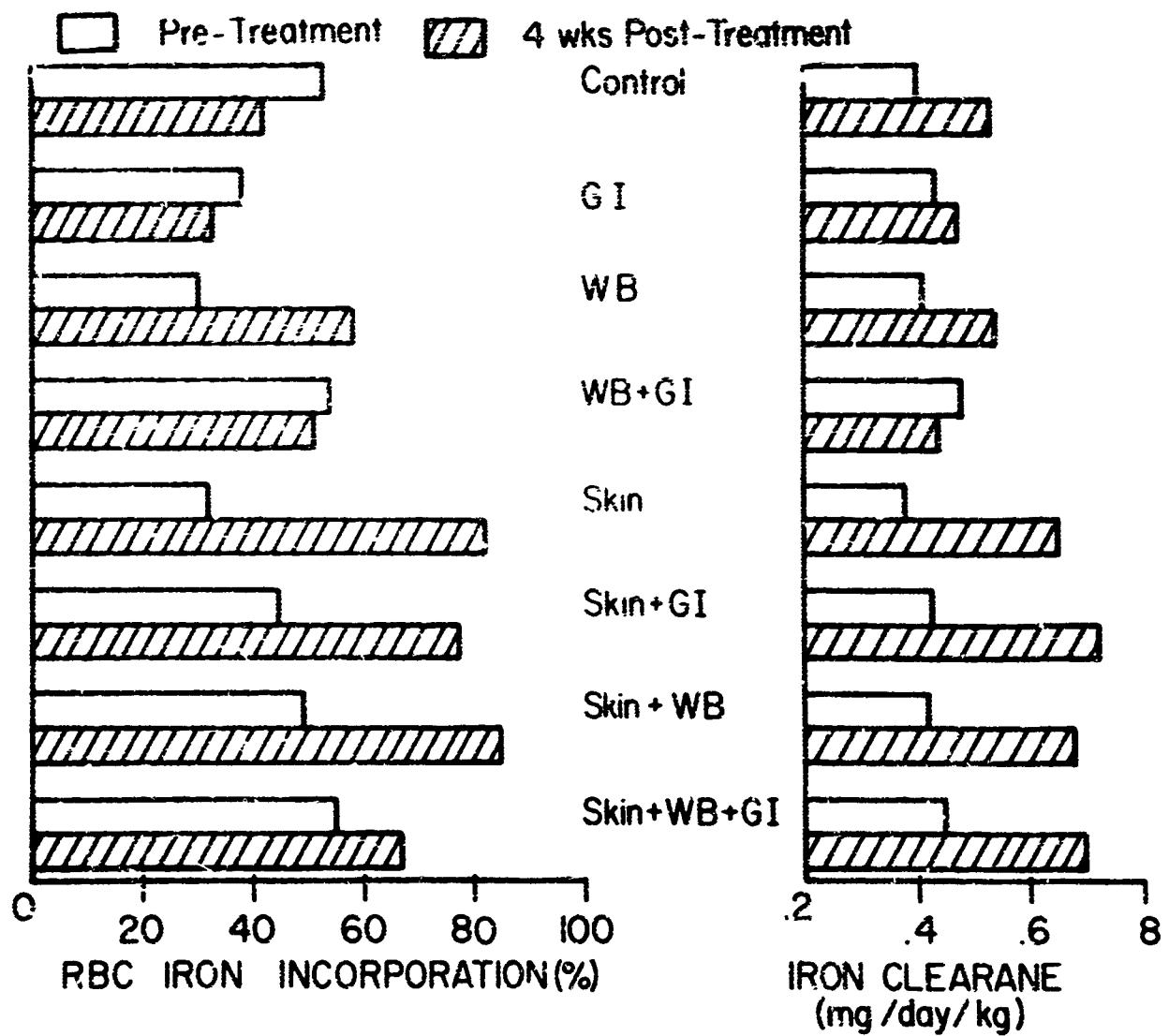


Figure 10. Effects of Irradiation on ^{59}Fe Clearance and Incorporation into Erythrocytes in Sheep

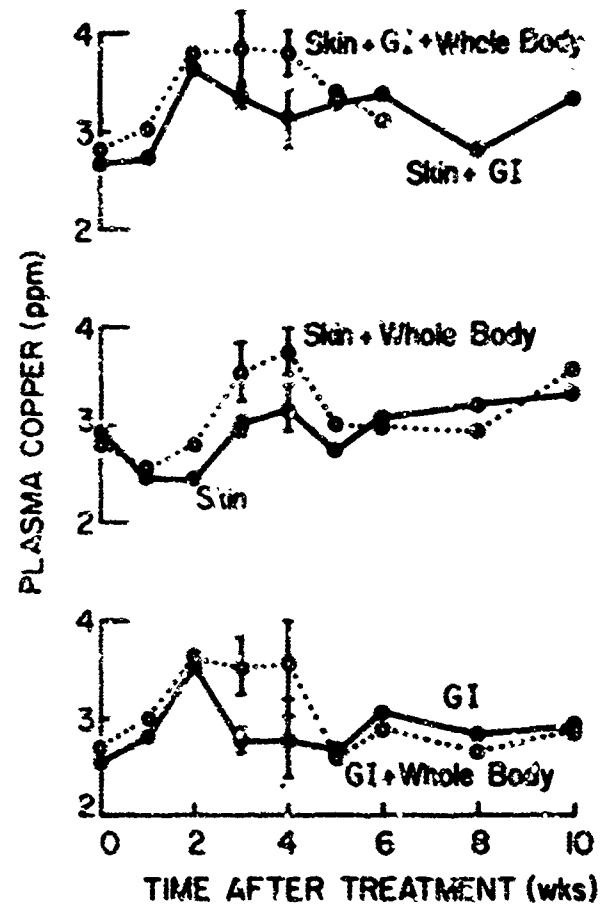
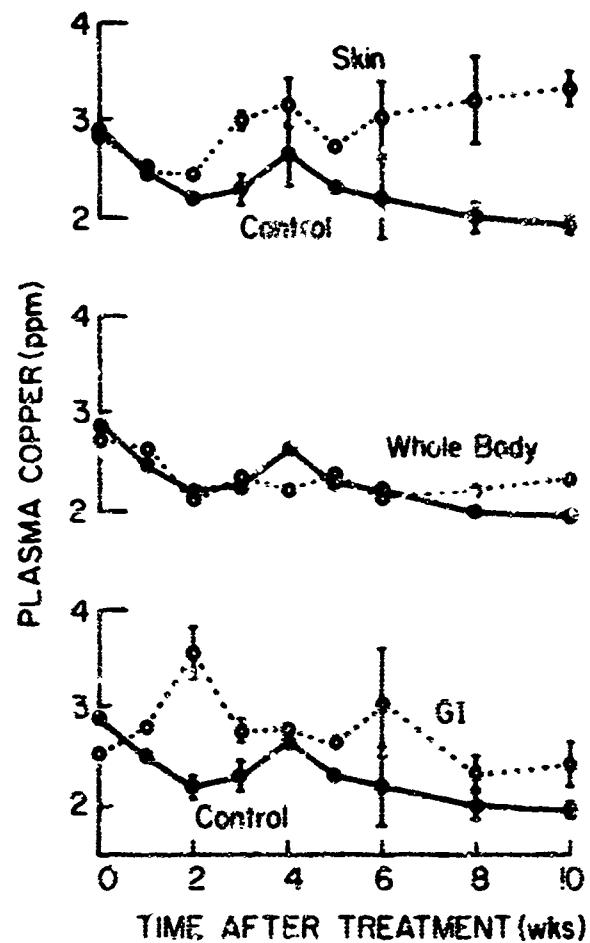


Figure 11. Effects of Irradiation on Plasma Cu in Sheep.

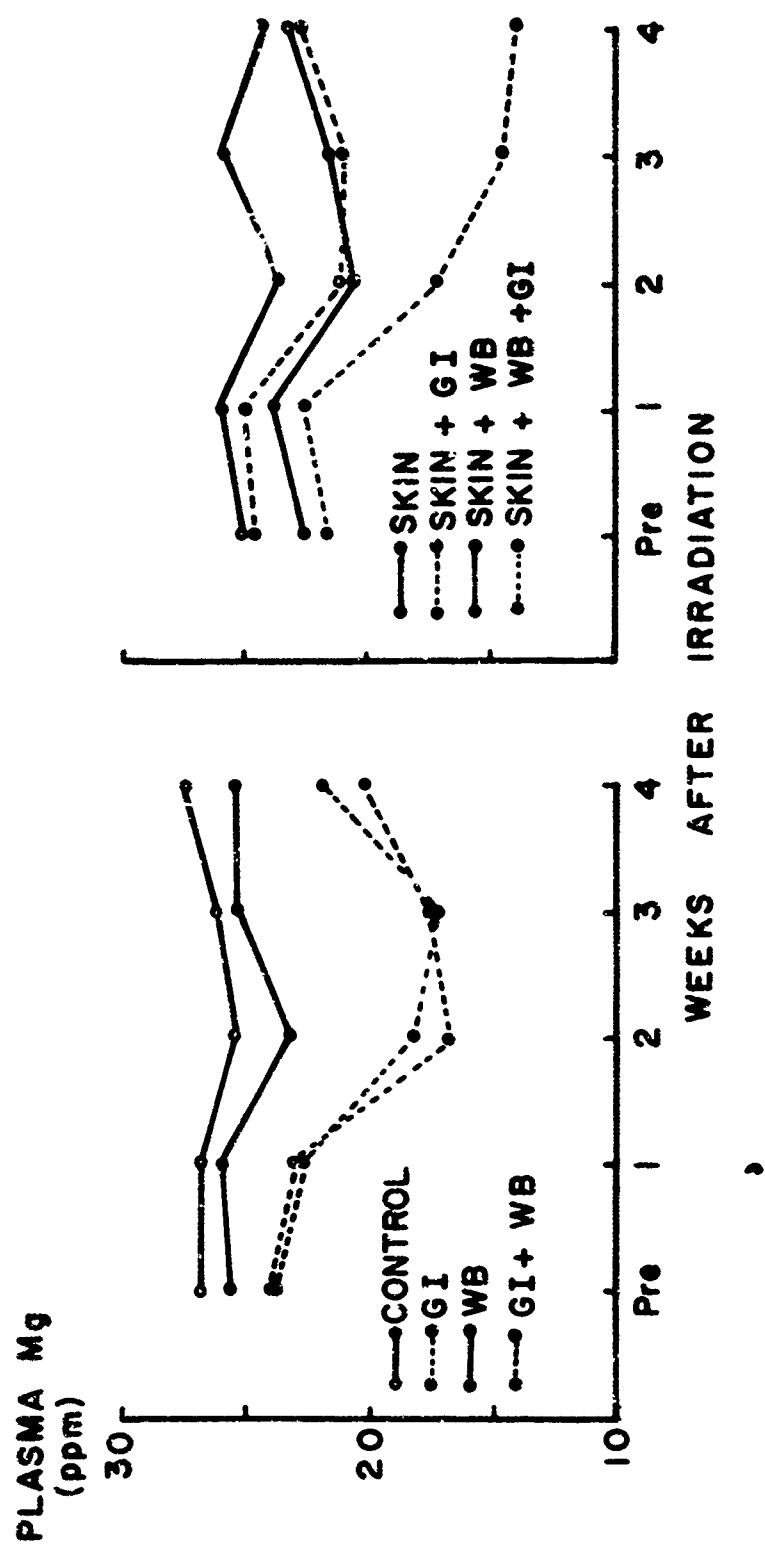


Figure 12. Effects of Irradiation on Plasma Mg in Sheep.

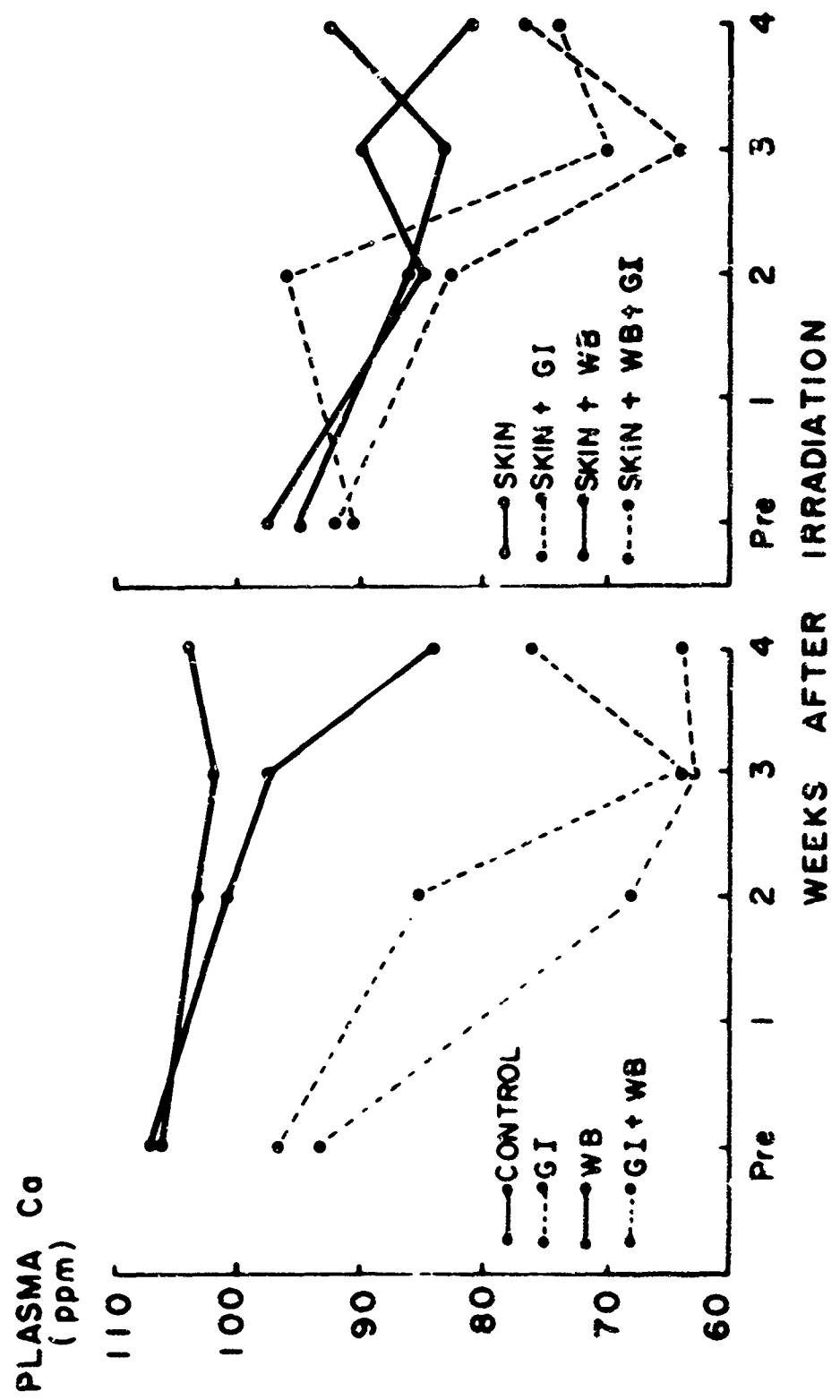


Figure 13. Effects of Irradiation on Plasma Ca in Sheep.

P A R T B.
FALLOUT RADIATION AND CROP PRODUCTIVITY
by
D. D. Killion and M. J. Constantin

ANNUAL REPORT
November 1, 1968 to October 31, 1969

Data on the extent of yield reduction in agronomic plants following sublethal levels of ionizing radiation are needed. Information available is generally derived from an experiment designed to fulfill other objectives, and is inadequate to permit valid estimation of potential damage to food producing capability of crop plants exposed to radioactive fallout contamination. Experiments conducted under work order number DAHC 20 were designed to assess the vulnerability of major agronomic plants to gamma radiation in lieu of radioactive fallout radiation. For the most part, experiments were designed to determine the extent of yield reduction following acute, sublethal exposures of gamma radiation to various plant species at different stages of growth (time after emergence).

The Variable Dose Rate Irradiation Facility (VDRIF), a ⁶⁰Co multiple source facility, was used. Plant species studied were soybeans (Glycine max. Merrill, var. Hill), winter barley (Hordeum vulgare L., var. Dayton), corn (Zea mays L., var. WF-9 x 38-11), winter wheat (Triticum vulgare, Vill, var. Seneca), and rice (Oryza sativa L., var. CI 8970-5). Plants were grown in containers (55-gal drums cut in half crosswise), irradiated at various times after emergence, and allowed to mature in the field. Some growth room studies were employed to supplement the field studies.

Stage sensitivity studies in 1968 on the above plant species have for the most part been completed, and are included in this report. Data for 1969 are not yet available, but observations indicate a pattern

of response similar to that observed in 1968. Studies on radiation factors, namely, exposure and exposure-rate of gamma radiation were initiated on some of the crops in 1969. These factors will be studied in more detail with the 1970 crop.

Rice. In the 1968 field study, 2500 R at 50 R/min administered from 1 to 25 days after emergence failed to affect grain production. Results of a growth room study showed that 20,000 R at 50 R/min produced some vegetative damage. Yield data for the 1969 field study are not yet available, but visual observations indicate that 25,000 R at 50 R/min produced vegetative damage and reduced grain yield. This study will provide in addition information on the extent of interaction between plant developmental stage and radiation exposure level.

Winter Barley. In growth room studies, winter barley seedlings were exposed to 250 to 2250 R in 250 R increments at 20 R/min one day after emergence. Dry weight and height of the seedlings were taken at the end of 3 weeks after emergence. Dry weight and height were reduced with increasing exposures of radiation, and maximal inhibition was obtained at 1250 and 1000 R, respectively. Tiller stimulation was evident in lower irradiation levels.

In the 1969 field study, the effect of radiation on winter barley at various times after emergence is shown in Fig. 1. Plants were exposed to 1600 R at 20 R/min. Grain production was noticeably reduced between 186 to 193 days after plant emergence. During this stage of plant development, spike initiation and emergence occurred. Some spikes did not emerge, indicating a failure of head formation or failure of peduncle cell division and elongation to lift the head above the flag

leaf. To compensate for shielding by the growth medium, young plants whose apical meristem was below the surface were exposed to 2200 R at 20 R/min. These plants did not survive the combined stress of winter conditions and irradiation damage.

Winter Wheat. Yield reduction of winter wheat grown in 1969 produced by irradiation is given in Fig. 2. Type of response was found to be similar to winter barley. Damage is most noticeable in the floral development stage, 160 to 200 days after emergence. The plants were exposed to 1600 R at 20 R/min.

Corn. Figure 3 shows that corn yield in 1968 was highly dependent upon time of irradiation. Plants were exposed to 2500 R at 50 R/min. No grain was produced on plants irradiated from 14 through 48 days after emergence. Shoot meristem in plants 1 to 13 days after emergence was below the growth medium surface and was thus partially shielded. Plants irradiated from 49 through 81 days after emergence showed increasing tolerance to gamma irradiation which is indicative of response of developing embryos. Yield data for the 1969 field study are not yet available; observations indicate a pattern response similar to that observed in the 1968 crop. This study also will provide information on the nature and extent of the interaction between development stage and radiation level.

Soybeans. Irradiation markedly influenced Hill soybean seed yield in 1968 as is shown in Fig. 4. Plant sensitivity declined until about 20 days after emergence, then increased to early bloom (21 to 46 days after emergence), after which sensitivity declined again. Data for 1969 indicate a response similar to that of 1968 for stage sensitivity.

In addition to stage sensitivity, exposure and exposure-rate of radiation are included in this study.

SUMMARY

Experiments have been conducted to determine the extent of yield reduction following acute, sublethal exposures of various crop plants at different stages of growth to gamma rays. The order of sensitivity to gamma rays for crop plants studied was as follows: winter barley = winter wheat > corn > soybean > rice. Seedlings of barley and wheat will tolerate ≈ 1 kR, corn ≈ 2 kR, soybean ≈ 4 kR, and rice ≈ 25 kR; however, the stage of growth influences the tolerance of plants to gamma rays. Maximum yield reduction occurred in corn irradiated from 14 to 48 DAE (days after emergence), in winter barley from 175 to 200 DAE, and in winter wheat from 150 to 200 DAE. Yield reduction in soybean was maximal at early seedling stages and during early bloom. In the monocotyledonous plants sensitivity to gamma rays is greatest during the period of apical transition from a vegetative to a reproductive apex and extends through the period of reproductive primordial development. In the one dicotyledonous species studied, radiosensitivity was related to the early postemergence stage when vegetative mass was being developed and during the period of early bloom.

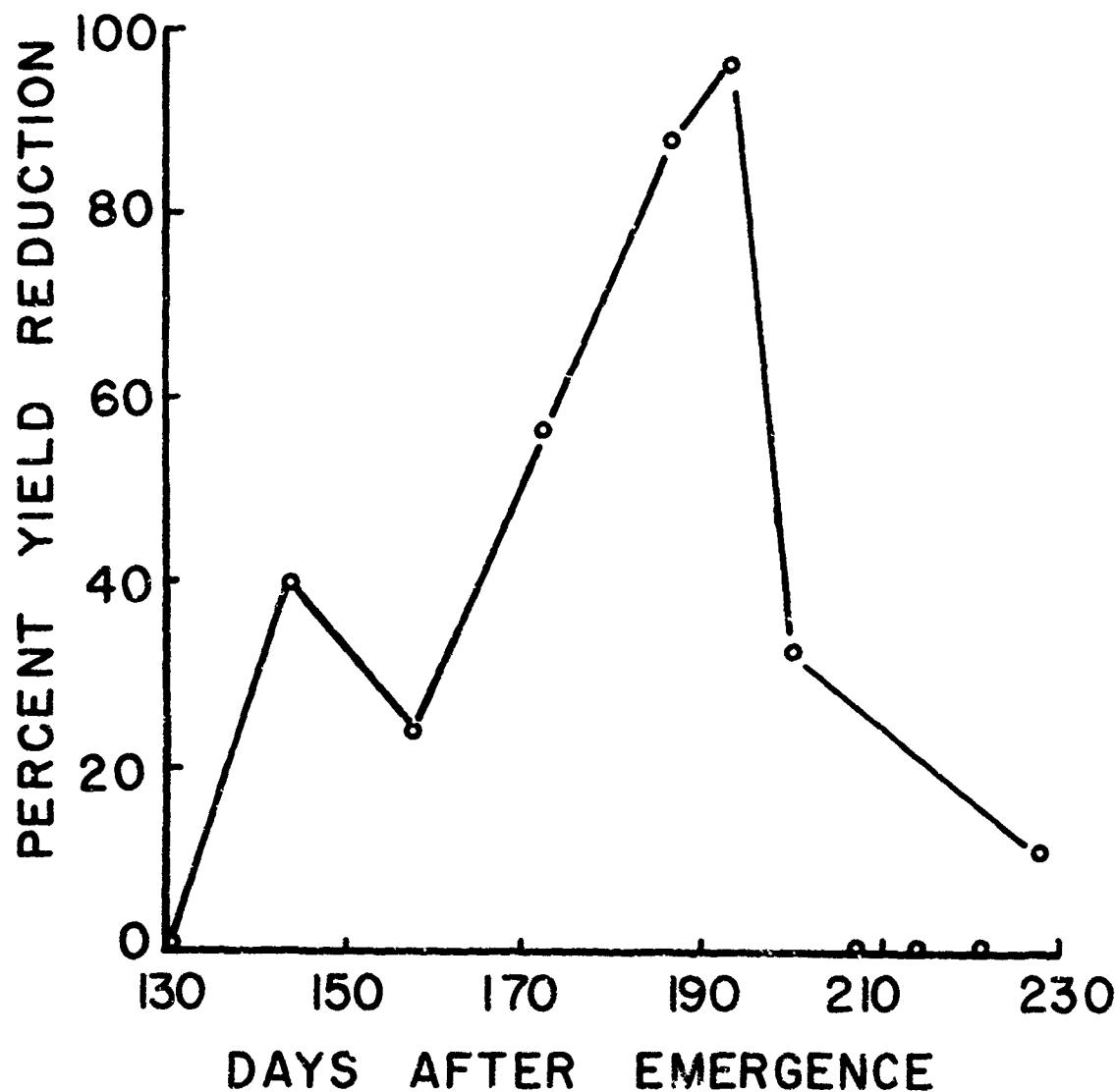


Figure 1. Grain Yield Reduction of Dayton Barley after Exposure to 1000 R at 20 R/min of ^{60}Co Gamma Radiation at Different Stages of Plant Growth (days after emergence). (Plants irradiated previous to 130 days after emergence did not survive winter conditions.)

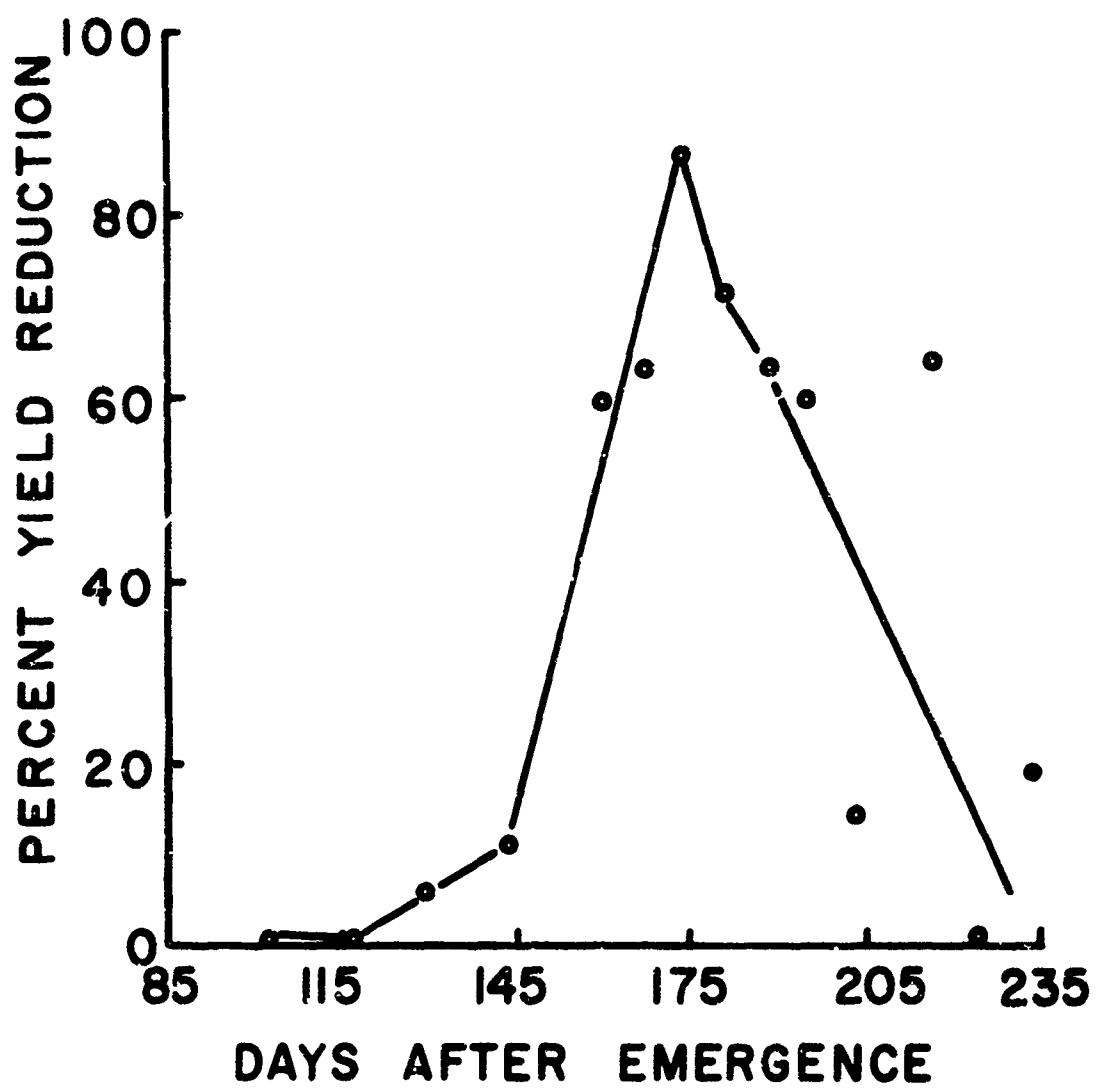


Figure 2. Grain Yield Reduction of Seneca Wheat after Exposure to 1600 R at 20 R/min of ^{60}Co Gamma Radiation at Different Stages of Plant Growth (days after emergence). (Plants irradiated previous to 85 days after emergence did not survive winter conditions.)

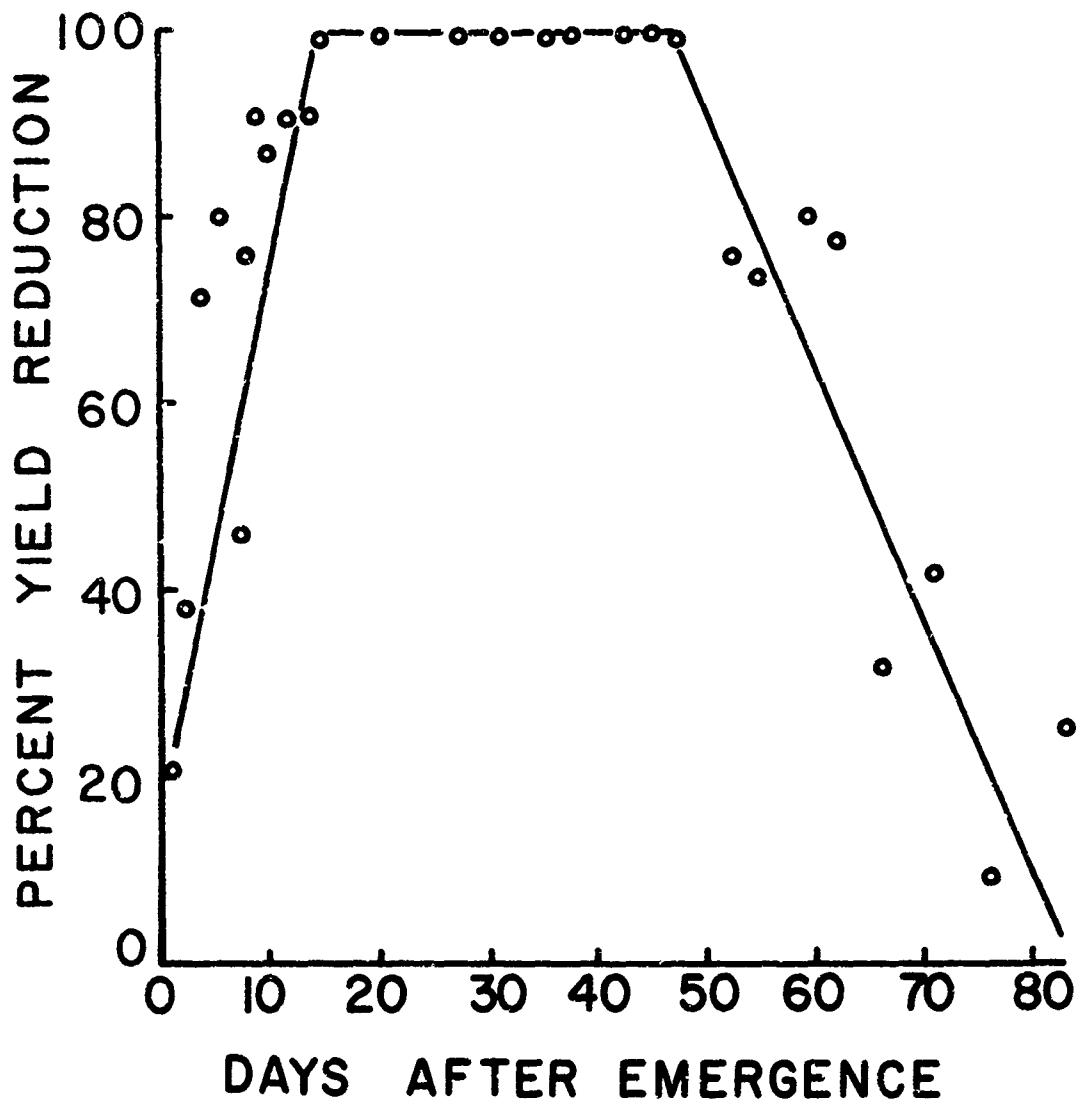


Figure 3. Grain Yield Reduction of WF-9 X 38-11 Corn after Exposure to 2500 R at 50 R/min of ^{60}Co Gamma Radiation at Different Stages of Plant Growth (days after emergence).

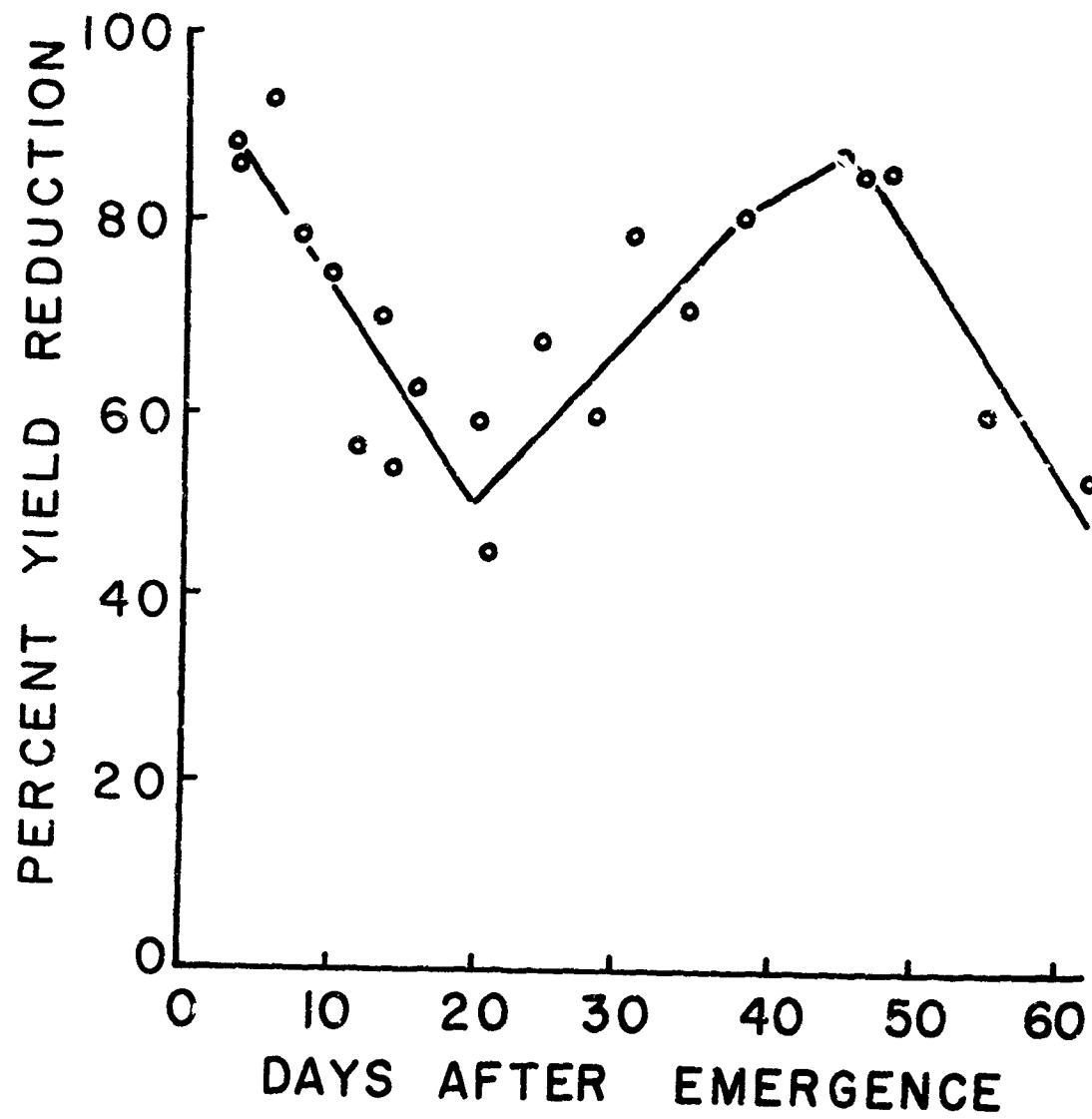


Figure 4. Seed Yield Reduction of Hill Soybeans after Exposure to 2500 R at 50 R/min of ^{60}Co Gamma Radiation at Different Stages of Plant Growth (days after emergence).

Unclassified

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Security Classification

Title: Fallout Radiation Effects on Livestock (Part A) and Food Crops (Part B)

Authors: M. C. Bell, L. B. Sasser, J. L. West, and L. Wade, Jr., (Part A); D. D. Killion and M. J. Constantin (Part B).

SUMMARY

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Fallout Simulant						

Security Classification